

MODELLING OF SOLID STATE FERMENTATION PROCESS

A DISSERTATION

*Submitted in partial fulfilment of the
requirements for the award of the degree
of*

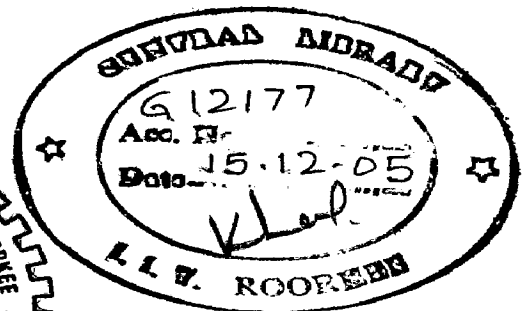
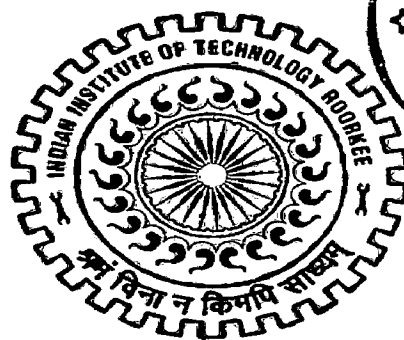
**MASTER OF TECHNOLOGY
in**

CHEMICAL ENGINEERING

(With Specialization in Computer Aided Process Plant Design)

By

ASAD HASAN SAHIR



**DEPARTMENT OF CHEMICAL ENGINEERING
INDIAN INSTITUTE OF TECHNOLOGY ROORKEE
ROORKEE-247 667 (INDIA)**

JUNE, 2005

CANDIDATE'S DECLARATION

I hereby certify that the work, which is being presented in the thesis entitled **“MODELLING OF SOLID STATE FERMENTATION PROCESS”** in partial fulfillment of the requirement for the award of the **Degree of Master of Technology in Chemical Engineering** with specialization in **Computer Aided Process Plant Design (CAPPD)**, and submitted in the **Department of Chemical Engineering of Indian Institute of Technology Roorkee**, is an authentic record of my own work carried out under the supervision of **Dr. Surendra Kumar**, Professor, Department of Chemical Engineering, Indian Institute of Technology Roorkee, Roorkee.

The matter presented in this **thesis** has not been submitted by me for the award of any other degree/diploma of this or any other Institute / University.

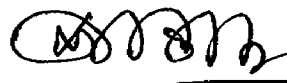
Date : June 29, 2005

Place : Roorkee



(Asad Hasan Sahir)

This is to certify that the above statement made by the candidate is correct to the best of my knowledge.



(Dr. Surendra Kumar)

Professor

Department of Chemical Engineering

Indian Institute of Technology, Roorkee

Roorkee - 247667

Date: June 29, 2005

ABSTRACT

Solid State Fermentation involves the fermentation of solid substrates at low water contents. The reduction of bioreactor volume, contamination-level, energy usage and separation costs associated with bioprocesses based on Solid State Fermentation have attracted huge amount of research interest in the past two decades.

Packed Bed Solid State Fermentation Bioreactors are one of the most commonly used bioreactor types in Solid State Fermentation processes, due to their simple design and constitution which allows efficient control of air flow rate and temperature.

In this thesis, mathematical models have been developed for Packed Bed Solid State Fermentation Bioreactors employing concepts envisaged in Chemical Reaction Engineering. The N-Tanks in Series approach has been utilized to analyze two different situations. The first situation involves the solids and air being considered as a single phase (pseudohomogeneous phase) and in the other situation; they are treated as distinct phases.

The primary importance of employing the N-Tanks in Series approach is to demonstrate its successful application to the solution of model equations which avoids the use of complex computational methods like Orthogonal Collocation. It also aids in providing more insights in the design of Packed Bed Solid State Fermentation Bioreactors.

The mathematical model developed for the pseudohomogeneous phase considers the production of protease by *Penicillium fellutanum* and *Aspergillus niger*. It demonstrates the threat posed by increased temperatures in packed beds to enzyme production and helps in proposing solutions to the problem. The developed mathematical model has been validated with the experimental data of Ghildyal et al. (1994). The mathematical model developed for the two – phase system aids in the exploration of the potential situations to which it can be applied.

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TABLE OF CONTENTS

ABSTRACT	i
ACKNOWLEDGEMENTS	ii
TABLE OF CONTENTS	iv
LIST OF FIGURES	ix
LIST OF TABLES	xv
NOMENCLATURE	xvi
CHAPTER 1 INTRODUCTION	1
1.1 SOLID STATE FERMENTATION	1
1.2 ADVANTAGES OF SOLID STATE FERMENTATION	1
1.3 CHALLENGES IN SOLID STATE FERMENTATION TECHNOLOGY	1
1.4 MICROBIOLOGICAL , ENVIRONMENTAL AND ENGINEERING ASPECTS OF SOLID STATE FERMENTATION TECHNOLOGY	2
1.4.1 Microbiological Aspects	2
1.4.2 Environmental Aspects	2
1.4.3 Engineering Aspects	2
1.5 BIOREACTOR DESIGNS IN SOLID STATE FERMENTATION	3
1.5.1 Challenges on Industrial Scale	3
1.5.2 Packed Bed Solid State Fermentation Bioreactor	4
1.5.2.1 The Zymotis Packed Bed Bioreactor Design	4

1.5.2.2	Reported Use of Packed Beds in Literature	6
1.6	INDUSTRIAL POTENTIAL OF SOLID STATE FERMENTATION TECHNOLOGY	7
1.7	REMARKS	8
1.8	OBJECTIVES OF THE THESIS	9
1.9	ORGANISATION OF THE THESIS	9
CHAPTER 2	LITERATURE REVIEW	10
2.1	MATHEMATICAL MODELS OF PACKED BED SOLID STATE FERMENTATION BIOREACTORS	10
2.2	MATHEMATICAL MODELS TREATING AIR AND SUBSTRATE BED AS A PSEUDOHOMOGENEOUS PHASE	11
2.2.1	Model by Saucedo-Castañeda(1990)	11
2.2.2	Model by Sangsurasak and Mitchell (1995)	12
2.2.3	Model by Sangsurasak and Mitchell (1998)	13
2.2.4	Model by Hasan et al. (1998)	15
2.2.5	Model by Mitchell et al. (1999)	16
2.2.6	Model by Ashley et al. (1999)	17
2.2.7	Model by Mitchell and Von Meien (2000)	18
2.2.8	Model by Mitchell et al. (2002)	20
2.2.9	Model by Dos Santos et al. (2004)	21
2.3	MATHEMATICAL MODELS TREATING AIR AND SUBSTRATE BED AS DISTINCT PHASES	22
2.3.1	Model of Weber et al. (1999)	22
2.3.2	Model of Weber et al. (2002)	23
2.3.3	Model of Von Meien and Mitchell (2002)	24
2.3.4	Model of Von Meien et al. (2004)	25

2.4	SIGNIFICANCE OF MATHEMATICAL MODELS	26
2.5	MOTIVATION FOR THE PRESENT STUDY	30
2.6	CONCLUDING REMARKS	31
CHAPTER 3	DEVELOPMENT OF THE MATHEMATICAL MODEL FOR A PACKED BED SOLID STATE FERMENTATION BIOREACTOR USING N-TANKS IN SERIES APPROACH	32
3.0	INTRODUCTION	32
3.1	MATHEMATICAL MODEL FOR A PSEUDOHOMOGENEOUS PHASE	32
3.1.1	Assumptions	33
3.1.2	Derivation of the Mathematical Model	33
3.1.3	Constitutive Relationships and Parameters Employed	39
3.1.3.1	Growth Kinetics	39
3.1.3.2	Enzyme Activity and Denaturation	40
3.1.3.3	Evaluation of Humidity	40
3.1.3.4	Geometric Considerations	40
3.1.3.5	Temperature Control Strategy	41
3.1.3.6	Parameters	42
3.1.4	Summary of Model Equations to be solved	42
3.1.5	Initial Conditions	42
3.1.6	Method of Solution and Computer Program	43
3.2	MATHEMATICAL MODEL FOR A TWO PHASE SYSTEM	44
3.2.1	Derivation of the Mathematical Model	44
3.2.2	Assumptions	44
3.2.3	Development of Model Equations	47
3.2.3.1	Mass Balance for water in air(gas phase)	47
3.2.3.2	Mass Balance for water in the dry solid (solid phase)	48

3.2.3.3	Equations for the production of biomass	50
3.2.3.4	Material Balance on Dry Solids	50
3.2.3.5	Energy Balance for air(gas phase)	51
3.2.3.6	Energy Balance for the Solid Phase	53
3.2.4	Constitutive Relationships	56
3.2.4.1	Initial water content for Air	56
3.2.4.2	Initial water content for Solids	56
3.2.4.3	Mass Transfer Coefficient	57
3.2.4.4	Heat Transfer Coefficient	57
3.2.4.5	Specific Growth Rate	57
3.2.5	Model Equations for the n th Bioreactor	58
3.2.5.1	Mass Balance for water in air(gas phase)	58
3.2.5.2	Mass Balance for water in the dry solid (solid phase)	58
3.2.5.3	Equations for the production of biomass	58
3.2.5.4	Material Balance on Dry Solids	58
3.2.5.5	Energy Balance for air(gas phase)	58
3.2.5.6	Energy Balance for the Solid Phase	59
3.2.6	Parameters for the solution of Mathematical Model	59
3.2.7	Initial Conditions	60
3.2.8	Method of Solution and Computer Program	61
3.3	CONCLUDING REMARKS	62
CHAPTER 4	RESULTS AND DISCUSSION	63
4.0	INTRODUCTION	63
4.1	RESULTS FROM THE PSEUDOHOMOGENEOUS MODEL	63
4.1.1	Profiles of Temperature, Solid Substrate Active Enzyme Content and Denatured Enzyme for Aspergillus niger	64

4.1.2	Profiles of Temperature, Solid Substrate Active Enzyme Content and Denatured Enzyme for <i>Penicillium fellutanum</i>	65
4.1.3	Effect of the gain factor in temperature control strategy	65
4.1.4	Study of the variation of Temperature, Solid Substrate Active Enzyme Content and Denatured Enzyme with height (axial distance)	66
4.2	RESULTS FROM THE TWO-PHASE MODEL	90
4.2.1	Computational Experiences	90
4.2.2	Profiles of temperature and water content of solid and gaseous phases, biomass and solid phase concentration with time	90
4.2.3	Comparison of the results from N-Tanks in Series approach with Von Meien and Mitchell (2002)	92
4.3	CONCLUDING REMARKS	112
 CHAPTER 5 CONCLUSIONS AND RECCOMENDATIONS		 113
5.1	CONCLUSIONS	113
5.1.1	Pseudohomogeneous Phase Model Developed by N-Tanks in Series approach	113
5.1.2	Two-Phase Model developed by N-Tanks in Series Approach	114
5.1.3	Final Remarks	115
5.2	RECOMMENDATIONS FOR FUTURE WORK	115
 REFERENCES		 116
APPENDIX A		123
APPENDIX B		125

LIST OF FIGURES

Figure 1.1:	Schematic diagram of a Packed Bed Solid State Fermentation Bioreactor system	5
Figure 3.1:	A schematic representing the system of bioreactors to be modelled as a pseudohomogeneous phase system	34
Figure 3.2:	A schematic representing the system of bioreactors to be modelled as a two phase system	46
Figure 4.1:	Plot of temperature vs. time for <i>Aspergillus niger</i> ($N=1, F_g =0, \mu=0.2, \alpha=0.2$)	68
Figure 4.2:	Plot of Solid State Active Enzyme Content vs. time for <i>Aspergillus niger</i> ($N=1, F_g =0, \mu=0.2, \alpha=0.2$)	68
Figure 4.3:	Plot of Denatured Enzyme vs. time for <i>Aspergillus niger</i> ($N=1, F_g =0, \mu=0.2, \alpha=0.2$)	69
Figure 4.4:	Plot of Biomass Content vs. time for <i>Aspergillus niger</i> ($N=1, F_g =0, \mu=0.2, \alpha=0.2$)	69
Figure 4.5:	Plot of temperature vs. time for <i>Penicillium fellutanum</i> ($N=1, F_g =0, \mu=0.0782, \alpha=0.0782$)	70
Figure 4.6:	Plot of Solid State Active Enzyme Content vs. time for <i>Penicillium fellutanum</i> ($N=1, F_g =0, \mu=0.0782, \alpha=0.0782$)	70
Figure 4.7:	Plot of Denatured Enzyme vs. time for <i>Penicillium fellutanum</i> ($N=1, F_g =0, \mu=0.0782, \alpha=0.0782$)	71
Figure 4.8:	Plot of Biomass Content vs. time for <i>Penicillium fellutanum</i> ($N=1, F_g =0, \mu=0.0782, \alpha=0.0782$)	71
Figure 4.9:	Plot of temperature vs. time for <i>Aspergillus niger</i> ($N=5, F_g =0, \mu=0.2, \alpha=0.2$)	72
Figure 4.10:	Plot of temperature vs. time for <i>Aspergillus niger</i> ($N=5, F_g =2, \mu=0.2, \alpha=0.2$)	72
Figure 4.11:	Plot of Solid State Active Enzyme Content vs. time for <i>Aspergillus niger</i> ($N=5, F_g =0, \mu=0.2, \alpha=0.2$)	73
Figure 4.12:	Plot of Solid State Active Enzyme Content vs. time for <i>Aspergillus niger</i> ($N=5, F_g =2, \mu=0.2, \alpha=0.2$)	73

- Figure 4.13: Plot of Denatured Enzyme vs. time for *Aspergillus niger* ($N=5$, $F_g=0$, $\mu=0.2$, $\alpha=0.2$) 74
- Figure 4.14: Plot of Denatured Enzyme vs. time for *Aspergillus niger* ($N=5$, $F_g=2$, $\mu=0.2$, $\alpha=0.2$) 74
- Figure 4.15: Plot of temperature vs. time for *Penicillium fellutanum* ($N=5$, $F_g=0$, $\mu=0.0782$, $\alpha=0.0782$) 75
- Figure 4.16: Plot of temperature vs. time for *Penicillium fellutanum* ($N=5$, $F_g=2$, $\mu=0.0782$, $\alpha=0.0782$) 75
- Figure 4.17: Plot of Solid State Active Enzyme Content vs. time for *Penicillium fellutanum* ($N=5$, $F_g=0$, $\mu=0.0782$, $\alpha=0.0782$) 76
- Figure 4.18: Plot of Solid State Active Enzyme Content vs. time for *Penicillium fellutanum* ($N=5$, $F_g=2$, $\mu=0.0782$, $\alpha=0.0782$) 76
- Figure 4.19: Plot of Denatured Enzyme vs. time for *Penicillium fellutanum* ($N=5$, $F_g=0$, $\mu=0.0782$, $\alpha=0.0782$) 77
- Figure 4.20: Plot of Denatured Enzyme vs. time for *Penicillium fellutanum* ($N=5$, $F_g=2$, $\mu=0.0782$, $\alpha=0.0782$) 77
- Figure 4.21: Plot of temperature vs. time for *Aspergillus niger* ($N=10$, $F_g=0$, $\mu=0.2$, $\alpha=0.2$) 78
- Figure 4.22: Plot of Solid State Active Enzyme Content vs. time for *Aspergillus niger* ($N=10$, $F_g=0$, $\mu=0.2$, $\alpha=0.2$) 78
- Figure 4.23: Plot of Denatured Enzyme vs. time for *Aspergillus niger* ($N=10$, $F_g=0$, $\mu=0.2$, $\alpha=0.2$) 79
- Figure 4.24: Plot of Biomass Content vs. time for *Aspergillus niger* ($N=10$, $F_g=0$, $\mu=0.2$, $\alpha=0.2$) 79
- Figure 4.25: Plot of temperature vs. time for *Penicillium fellutanum* ($N=10$, $F_g=0$, $\mu=0.0782$, $\alpha=0.0782$) 80
- Figure 4.26: Plot of Solid State Active Enzyme Content vs. time for *Penicillium fellutanum* ($N=10$, $F_g=0$, $\mu=0.0782$, $\alpha=0.0782$) 80
- Figure 4.27: Plot of Denatured Enzyme vs. time for *Penicillium fellutanum* ($N=10$, $F_g=0$, $\mu=0.0782$, $\alpha=0.0782$) 81

Figure 4.28:	Plot of Biomass Content vs. time for <i>Penicillium fellutanum</i> ($N=10, F_g=0, \mu=0.0782, \alpha=0.0782$)	81
Figure 4.29:	Plot of temperature vs. length of bioreactor for <i>Aspergillus niger</i> ($F_g=0, \mu=0.2, \alpha=0.2$)	82
Figure 4.30:	Plot of Solid State Active Enzyme Content vs. length of bioreactor for <i>Aspergillus niger</i> ($F_g=0, \mu=0.2, \alpha=0.2$)	82
Figure 4.31:	Plot of Denatured Enzyme vs. length of bioreactor for <i>Aspergillus niger</i> ($F_g=0, \mu=0.2, \alpha=0.2$)	83
Figure 4.32:	Plot of temperature vs. length of bioreactor for <i>Penicillium fellutanum</i> ($F_g=0, \mu=0.0782, \alpha=0.0782$)	83
Figure 4.33:	Plot of Solid State Active Enzyme Content vs. length of bioreactor for <i>Penicillium fellutanum</i> ($F_g=0, \mu=0.0782, \alpha=0.0782$)	84
Figure 4.34:	Plot of Denatured Enzyme vs. length of bioreactor for <i>Penicillium fellutanum</i> ($N=10, F_g=0, \mu=0.0782, \alpha=0.0782$)	84
Figure 4.35:	Validation of the N-Tanks in Series Model with the data of Ghildyal et al. (1994) for a flow rate of 5 lpm for <i>Aspergillus niger</i>	87
Figure 4.36:	Validation of the Two Dimensional Model (Sangsurasak and Mitchell,1998) with the data of Ghildyal et al. (1994) for a flow rate of 5 lpm for <i>Aspergillus niger</i>	87
Figure 4.37:	Validation of the N-Tanks in Series Model with the data of Ghildyal et al. (1994) for a flow rate of 15 lpm for <i>Aspergillus niger</i>	88
Figure 4.38:	Validation of the Two Dimensional Model (Sangsurasak and Mitchell,1998) with the data of Ghildyal et al. (1994) for a flow rate of 15 lpm for <i>Aspergillus niger</i>	88
Figure 4.39:	Validation of the N-Tanks in Series Model with the data of Ghildyal et al. (1994) for a flow rate of 15 lpm for <i>Aspergillus niger</i>	89
Figure 4.40:	Validation of the Two Dimensional Model (Sangsurasak and Mitchell,1998) with the data of Ghildyal et al. (1994) for a flow rate of 15 lpm for <i>Aspergillus niger</i>	89
Figure 4.41:	Plot of temperature of solid phase vs. time for $N=1$	95

Figure 4.42:	Plot of water content of solid phase vs. time for N=1	95
Figure 4.43:	Plot of concentration of total dry solids vs. time for N=1	96
Figure 4.44:	Plot of biomass concentration vs. time for N=1	96
Figure 4.45:	Plot of air(gas phase) temperature vs. time for N=1	97
Figure 4.46:	Plot of moisture content of air (gas phase) vs. time for N=1	97
Figure 4.47:	Plot of temperature of solid phase vs. time for N=3	98
Figure 4.48:	Plot of water content of solid phase vs. time for N=3	98
Figure 4.49:	Plot of solid phase concentration vs. time for N=3	99
Figure 4.50:	Plot of biomass concentration vs. time for N=3	99
Figure 4.51:	Plot of gas phase(air) temperature vs. time for N=3	100
Figure 4.52:	Plot of gas phase(air) water content vs. time for N=3	100
Figure 4.53:	Plot of temperature of solid phase vs. time for N=5	101
Figure 4.54:	Plot of water content of solid phase vs. time for N=5	101
Figure 4.55:	Plot of solid phase concentration vs. time for N=5	102
Figure 4.56:	Plot of biomass concentration vs. time for N=5	102
Figure 4.57:	Plot of gas phase(air) temperature vs. time for N=5	103
Figure 4.58:	Plot of gas phase(air) water content vs. time for N=5	103
Figure 4.59:	Plot of temperature of solid phase vs. axial distance at t=4.5 hours as determined by N-Tanks in Series model at which the solids temperature at the outlet is near maximal.	104
Figure 4.60:	Plot of temperature of solid phase vs. axial distance at t=12 hours as determined by Von Meien and Mitchell(2002)at which the solids temperature at the outlet is near maximal.	104
Figure 4.61:	Plot of temperature of gas phase vs. axial distance at t=4.5 hours as determined by N-Tanks in Series model at which the solids temperature at the outlet is near maximal.	105

Figure 4.62:	Plot of temperature of gas phase vs. axial distance at t=12 hours as determined by Von Meien and Mitchell(2002)at which the solids temperature at the outlet is near maximal.	105
Figure 4.63:	Plot of water activity of solid phase vs. axial distance at t=4.5 hours as determined by N-Tanks in Series model at which the solids temperature at the outlet is near maximal.	106
Figure 4.64:	Plot of water activity of solid phase vs. axial distance at t=12 hours as determined by Von Meien and Mitchell(2002)at which the solids temperature at the outlet is near maximal.	106
Figure 4.65:	Plot of water activity of gas phase vs. axial distance at t=4.5 hours as determined by N-Tanks in Series model at which the solids temperature at the outlet is near maximal.	107
Figure 4.66:	Plot of water activity of gas phase vs. axial distance at t=12 hours as determined by Von Meien and Mitchell(2002)at which the solids temperature at the outlet is near maximal.	107
Figure 4.67:	Plot of Biomass Concentration vs. time at the mid-height of bed as determined by N-Tanks in Series Model	108
Figure 4.68:	Plot of Biomass Concentration vs. time at the mid-height of bed as determined by Von Meien and Mitchell(2002)	108
Figure 4.69:	Plot of Water content of solid phase vs. time at the mid-height of bed as determined by N-Tanks in Series Model	109
Figure 4.70:	Plot of Water content of solid phase vs. time at the mid-height of bed as determined by Von Meien and Mitchell(2002)	109
Figure 4.71:	Plot of temperature of solid phase vs. time for the correlation cited in Mancini et al. as determined by N-Tanks in Series Model	110
Figure 4.72:	Plot of water content of solid phase vs. time for the correlation cited in Mancini et al. as determined by N-Tanks in Series Model	110
Figure 4.73:	Plot of solid phase concentration vs. time for the correlation cited in Mancini et al. as determined by N-Tanks in Series Model	110
Figure 4.74:	Plot of temperature of gaseous phase vs. time for the correlation cited in Mancini et al. as determined by N-Tanks in Series Model	111

Figure 4.75: Plot of water content of gaseous phase vs. time for the correlation cited in Mancini et al. as determined by N-Tanks in Series Model 111

Figure 4.76: Plot of biomass concentration vs. time for the correlation cited in Mancini et al. as determined by N-Tanks in Series Model 111

LIST OF TABLES

Table 1-1:	Examples of usage of Packed Bed Solid State Fermentation Bioreactors reported in literature.	6
Table 2-1:	Significance of Mathematical Models for Packed Bed Solid State Fermentation Bioreactors.	27
Table 3-1:	Various parameters and their values employed for the solution of model based on pseudohomogeneous phase system	41
Table 3-2:	Various parameters and their values employed for the solution of a model based on a two-phase system.	59

NOMENCLATURE

a_1, a_2, a_3, a_4	=	Constants for the evaluation of mass transfer coefficient
a_{wg}	=	Water activity of the gas phase
a_{ws}	=	Water activity of the solid phase
A	=	Area of cross section (m^2)
A_0	=	Frequency factor for denaturation (h^{-1})
A_T	=	Frequency factor for evaluation of fractional specific growth rate (s^{-1})
b	=	Concentration of biomass (kg biomass/kg total dry solids).
b_m	=	Maximum possible biomass concentration (kg biomass/kg total dry solids)
B	=	Total mass of solids in the substrate bed (kg)
B_T	=	Frequency factor for the evaluation of fractional specific growth rate (s^{-1}).
c_1, c_2, c_3, c_4, c_5	=	Constants for the evaluation of water activity of the solid phase.
C_p	=	Specific Heat (J/kg K)
D	=	Diameter of bed (m)
d_1, d_2, d_3, d_4	=	Constants for the evaluation of specific growth rate based on the water activity
D_e	=	Denatured Enzyme (PU/g IDS)
E	=	Solid Substrate Active Enzyme Content (PU/g IDS)
E_{A1}, E_{A2}	=	Activation energies in the equation for the calculation of fractional specific growth rate based on temperature (J/mol)
E_m	=	Solid Substrate Maximum Enzyme Content (PU/g IDS)
E_{n_a}	=	Denaturation Reaction Activation Energy (J/mol)
F	=	Mass flow rate (kg/s)
F_g	=	Gain factor of temperature control strategy mechanism
G	=	Mass flow rate of air per unit cross sectional area (kg/m^2s)

h	=	Bioreactor Wall Heat Transfer Coefficient ($J h^{-1} m^2 ^\circ C$)
h_a	=	Humidity of air (kg vapor/kg dry air)
h_t	=	Coefficient for heat transfer between gas and dry solid bed ($J s^{-1} m^{-3} K$)
H'_S	=	Enthalpy of the system(J)
H	=	Height of the bioreactor (m)
\underline{H}'_S	=	Specific Enthalpy (J/kg)
k	=	Denaturation reaction rate constant (h^{-1})
K_a	=	Mass transfer coefficient for water transfer between solid and gas phase ($kg H_2O s^{-1} m^{-3}$)
KE	=	Kinetic Energy of the system (J)
\underline{KE}	=	Kinetic Energy per unit mass (J/kg)
L	=	Length of the bioreactor (m)
P	=	Pressure (N/m^2)
P_{vap}	=	Water vapor pressure at air outlet (mm Hg)
p_w^{sat}	=	Saturation vapor pressure in the gas phase (N/m^2)
PE	=	Potential Energy of the system (J)
\underline{PE}	=	Potential Energy of the system per unit mass (J/Kg)
q	=	Volumetric flow rate(m^3/s)
Q	=	Heat transferred (J)
R	=	Universal gas constant ($J/mol ^\circ C$)
S	=	Dry Solid Concentration (kg/m^3)
t	=	Time
T	=	Temperature ($^\circ C$)
T_s	=	Temperature of control strategy point ($^\circ C$)
U	=	Internal energy of the system (J)
\underline{U}	=	Internal energy per unit mass (J/kg)
V	=	Volume of the system (m^3)
W	=	Water Content of the bed (kg water /kg initial dry solids)
W_s	=	Shaft Work (J)
W_T	=	Total Work (J)

X	=	Biomass content of the solid substrate (kg biomass/kg IDS)
X_m	=	Maximum possible biomass content (kg biomass/kg IDS)
Y_Q	=	Heat Yield from growth (J/kg biomass)
Y_{SB}	=	Change in total dry solids per kg of biomass produced (kg total dry solids/kg biomass)
Y_{WB}	=	Stoichiometric Coefficient relating water production to growth (kg water/kg biomass)

Greek Symbols:-

α	=	Enzyme production specific rate constant (h^{-1})
Δ	=	It represents change in time, water content and temperature.
ε	=	Void Fraction
λ	=	Enthalpy of vaporization (J kg vapor ⁻¹)
μ	=	Specific growth rate (time ⁻¹)
μ_{opt}	=	Optimum specific growth rate (time ⁻¹)
μ_T	=	Fractional specific growth rate based on the substrate bed temperature
μ_W	=	Fractional specific growth rate based on substrate bed water activity
ρ	=	Density (kg/m ³)
ρ_g	=	Density of air (kg/m ³)
Φ_s	=	Water content of solid phase (kg water /kg total dry solids)
Φ_s^*	=	Solid phase water content for equilibrium with the gas phase at air temperature (kg water / kg dry solids)

Subscripts:-

a	=	Air at entry to the first mixed tank
b	=	Moist Substrate Bed
e_n	=	Exit of the n^{th} mixed tank ($n = 1, 2, \dots$)
f_n	=	Entry to the n^{th} mixed tank ($n = 1, 2, \dots$)
g	=	Gaseous Phase(Dry Air)
g_n	=	Air in the n^{th} bed ($n = 1, 2, \dots$)
o	=	Initial contents (Biomass, Solid Substrate Active Enzyme; Denatured Enzyme)
ref	=	Reference
S	=	Dry Solid
Sh	=	Shaft Work
S_n	=	Substrate in the n^{th} mixed tank
T	=	Temperature
v	=	Water vapor
w	=	Water

Abbreviations:-

IDS	=	Initial dry solids
PU	=	Protease Unit
SSF	=	Solid State Fermentation (Solid Substrate Fermentation)

INTRODUCTION

1.1 SOLID STATE FERMENTATION

Solid State Fermentation (SSF) is a microbial process in which a solid material is used as the substrate or the inert support on which microorganisms grow (Sato and Sudo, 1999).

The solid porous matrix should be porous, uncontaminated, possess a large surface area per unit volume and have a relatively high water activity (Ratio of equilibrated partial vapor pressure over the substrate and pure water) assuring high biochemical reaction rates.

1.2 ADVANTAGES OF SOLID STATE FERMENTATION

Solid State Fermentation has the following advantages:

1. It is relatively resistant to bacterial contamination.
2. The fermentative facilities employed are compact than submerged fermentation.
3. Extraction of product in the process requires less solvent.
4. Simple treatment of fermented residue is possible (Robinson and Nigam, 2003).

1.3 CHALLENGES IN SOLID STATE FERMENTATION TECHNOLOGY

The engineering challenges in Solid State Fermentation can be attributed to the heterogeneity of physiological, physical and chemical environment in the substrate bed, which makes the rapid determination of microbial growth difficult.

Since the solid substrate bed has a very low thermal conductivity, hence temperature control of heat evolved by microbial respiration or metabolism is difficult (Paredes-López et al., 1998).

1.4 MICROBIOLOGICAL, ENVIRONMENTAL AND ENGINEERING ASPECTS OF SOLID STATE FERMENTATION TECHNOLOGY

1.4.1 Microbiological Aspects

The hyphal mode of fungal growth and their excellent tolerance to variations in water activity, make fungi the most suitable species. High water activities favour sporulation and low water activities favour mycelial growth. (Raimbault, 1998)

Substrates composed of lignocelluloses; pectins and starch are used in Solid State Fermentation processes.

1.4.2 Environmental Aspects

Solid State Fermentation processes are best suited for those microorganisms which are capable of carrying out their metabolic activities at low water activity. The moisture content should be optimal, a very low moisture content could lead to a hindrance in growth and an excess might result in anaerobiosis. The pH of the substrate should be preferably in the range of 5-8.

1.4.3 Engineering Aspects

The engineering aspects in Solid State Fermentation, (Raghavarao et al., 2003) pertain primarily to heat and mass transfer effects. The mass transfer processes which occur could be categorized as microscale and macroscale phenomena. Diffusion of oxygen into the substrate bed and degradation of the solid substrate by the diffusing enzymes constitute the primary intraparticle mass transfer effects (Rajgopalan and Modak, 1995). The bulk flow of air into and out of the bioreactor and the changes in O₂, CO₂ concentrations are the macroscale mass transfer effects.

In a Solid State Fermentation process, considerable amount of heat is evolved as a result of metabolic activities of microorganisms. It results in the development of steep temperature gradients. The temperature of substrate is a critical parameter, since a high temperature will result in spore germination,

growth and product formation, but simultaneously would cause drying in the substrate bed, a deleterious effect. A low temperature does not favour growth and other biochemical reaction rates.

Agitation of the fermenting mass is an important feature which provides new surfaces to aeration, inoculum distribution, promotes homogeneity and prevents aggregate formation. Intense agitation is not possible since it may cause damage to the hyphae of fungi involved in the process. The particle size of the substrate plays a crucial role; a smaller size provides a large specific surface area which is advantageous for growth of mould and facilitates heat transfer and exchange of CO₂ and O₂; but if it is too small then it would cause particles to adhere with each other.

1.5 BIOREACTOR DESIGNS IN SOLID STATE FERMENTATION

The different types of bioreactors used in Solid State Fermentation processes at the industrial scale are – Tray type (Koji type), Rotary drum fermentors (Continuously/Discontinuously Rotating), Packed Bed Solid State Fermentation Bioreactors. (Durand, 2003)

1.5.1 Challenges to Industrial Scale Usage

According to Durand (2003), the following are the major challenges encountered in the use of Solid State Fermentation Bioreactors at the industrial scale:-

1. Heat and mass transfer is affected by the difficulty in heat removal from the substrate bed.
2. Agitation causes destruction to fungal hyphae.
3. The choice of substrate, its possibilities for pre-treatment and its acceptable sterility level are the important issues to be addressed, during the development of any solid state fermentation process.

1.5.2 Packed Bed Solid State Fermentation Bioreactor

The basic design feature of a Packed Bed Solid State Fermentation Bioreactor (Figure 1-1) is the introduction of air through a sieve which supports the substrate. The reactor has to be steam sterilized and should have a scheme which facilitates control of inlet air temperature, water addition and agitation. The packed bed bioreactor can also be useful other than its use in industry in the following respects:-

1. Heat and mass transfer studies.
2. Provision of insights in the absence of mathematical models.
3. Determination of environmental parameters for temperature and substrate bed moisture.

The pressure drops are generally of the order of 0.1 to 0.5 cm. water per cm. of bioreactor height, although values as high as 1.4 to 2.7 cm. water per cm. of bioreactor height has also been observed in Packed Bed Solid State Fermentation Bioreactors.

1.5.2.1 The Zymotis Packed Bed Reactor Design

The Zymotis Bioreactor was developed by the ORSTOM team in France. It is a rectangular packed-bed bioreactor, aerated from the bottom with moist air. During the process the substrate bed remains static. For preventing the development of temperature gradients from front to back, an outer casing of insulated material is provided. It consists of vertical heat transfer plates in which cold water circulates. In between the plates; the previously inoculated medium is placed. The ORSTOM team had produced *Trichoderma harzianum* on 13 -40 kg of sugarcane bagasse and wheat bran mixture in a 100 l Zymotis Bioreactor (0.5m.long, 0.4 m. wide by 0.65 m. high) .

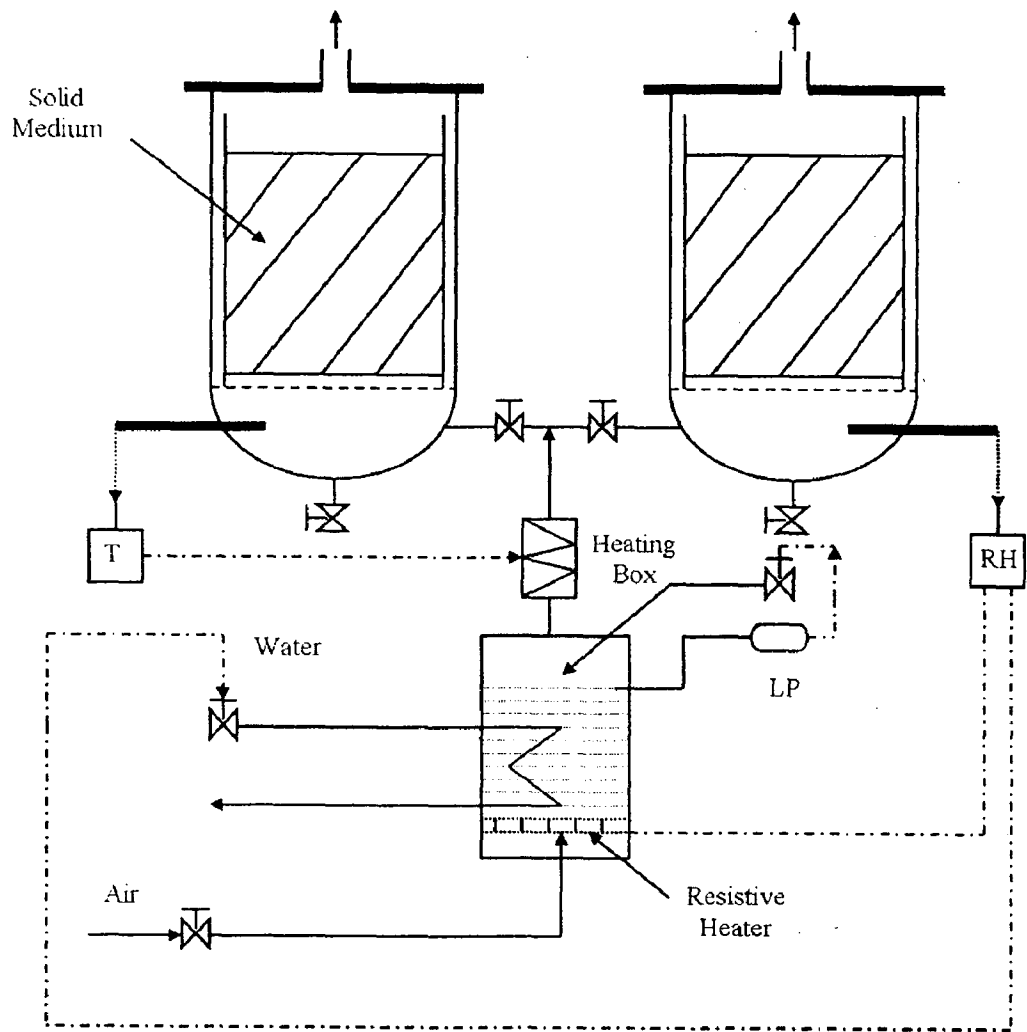


Figure 1.1 Schematic diagram of a Packed Bed Solid State Fermentation Bioreactor system.

1.5.2.2 Reported Use of Packed Beds in Literature

Mitchell et al., (2000) have mentioned the use of packed bed bioreactor for the following processes which have been experimentally reported in literature.

Table-1-1 Examples of usage of Packed Bed Solid State Fermentation Bioreactors reported in literature

Product	Species used for production	Reactor Configuration
Ethanol	<i>Schwanniomyces castelli</i>	4. 1 kg packed bed of nutrient enriched bagasse, 64 cm. long and 15 cm. diameter bed, aerated only for the first 10 hour of operation.
Oxytetracycline	<i>Streptomyces rimosus</i>	Production with a sweet potato residue in a packed bed, 14. 7 cm. in diameter and 50 cm. high.
Cellulase	<i>Trichoderma viride</i>	Production with a chaff and wheat bran mixture. Substrate contained within baskets (totalling 20 l) held within a cylinder 2 m long and 0. 5 m in diameter. Forced aeration achieved by periodical variation of

		air pressure within the vessel.
Citric Acid	Aspergillus niger	1. 5 kg (wet weight) Amberlite imbibed with nutrients within a column of 9. 5 cm. diameter and 25 cm. height, placed within a temperature controlled chamber.

1.6 INDUSTRIAL POTENTIAL OF SOLID STATE FERMENTATION TECHNOLOGY

Solid State Fermentation (SSF) Technology has tremendous industrial potential. Pandey et al.,(2000) have reviewed the production of bioactive products like mycotoxins, gibberellins; organic acids like citric and lactic acid and entomopathogenic and mycoparasitic fungi as biopesticides. They have also discussed the importance of SSF in the development of bioprocesses for biopulping, biological detoxification and biotransformation of crop residues and biodegradation and bioremediation of hazardous compounds. The potential of using SSF for the production of Ethanol is also being explored. The SSF Ethanol process is attractive due to its high volumetric productivity and the potential of the solid wastes or biomass generated in the process to be used as the substrate for another SSF operation. Ethanol has been produced using natural materials such as beets, fruit pomace, sweet sorghum corn, cassava or other tubers. The challenges for ethanol production include fermenter design and the moisture content which restricts ethanol production (Sato and Sudo, 1999). Many antibiotics like penicillin, cephalosporins, tetracyclines, cyclosporine and other secondary metabolites e.g. ergot alkaloids and gibberelic acid can be produced by SSF. The production of

surfactin (a lipopeptide antibiotic which acts as an inhibitor of fibrin clotting and erythrocyte lysing) was four to five times higher than submerged fermentation (Balakrishnan and Pandey, 1996).

Biocon India a Bangalore based company (Suryanarayan, 2003) envisaged the solid substrate fermentation technology originally for the production of enzymes. The company has been successful in producing industrial enzymes like glucoamylase, cellulose, tannase, pectinase, xylanase, lipases, phytase as well as microbial rennet. The enzymes produced by SSF were richer in side activities compared to commercially available submerged fermentation enzymes, when normalized to the same normal activity. For example a betaglucanase enzyme useful for the brewing industry had 17% xylanase when produced by solid state fermentation but when the same strain was produced by submerged fermentation, the xylanase percentage was 12%(30% lower). A plant in February 2000 based upon the PlaFactor[®] Technology (Mazumdar –Shaw and Suryanarayanan, 2003) and several different kinds of processes have been successfully demonstrated.

These include the production of lovastatin (an antihypercholesteremic agent), mycophenolic acid (an immunosuppressant), *Trichoderma viridae* spores (for biocontrol purposes) as well as a fungal protease as well as a fungal protease (enzyme for the food industry).

1.7 REMARKS

Solid State Fermentation can be employed for low cost and high volumetric production of enzymes, secondary metabolites and various chemicals. The heterogeneities associated at various scales with the phenomena present engineering challenges which have to be successfully addressed for the proper implementation of any bioprocess based upon this phenomenon.

1.8 OBJECTIVES OF THE THESIS

The main objectives of the thesis are:-

a) Development of Mathematical Model

To develop a mathematical model of a Packed Bed Solid State Fermentation Bioreactor using the N-Tanks in Series approach for the solid and gaseous phase in the packed bed considered as a single phase (pseudohomogeneous phase) or as a two-phase system.

b) Numerical Solution of the Model Equations

- i) To obtain the numerical solution of the model equations for specified sets of input data.
- ii) To explore the potential of the use of the N-Tanks in Series approach in the design of Packed Bed Solid State Fermentation Bioreactors and aim to reduce the complexities in computation involved.

c) Validation of the Proposed Model

To validate the mathematical model with experimental data and to analyse model predictions vis-à-vis results reported in literature.

1.9 ORGANISATION OF THE THESIS

The thesis has been organized into five chapters. In Chapter 2, the various mathematical models for Packed Bed Solid State Fermentation Bioreactors proposed till date have been reviewed. In Chapter 3, the mathematical model employing the N-Tanks in Series approach has been developed using mass and energy conservation laws. It also describes the requisite constitutive relationships and the strategy adopted for the solution of model equations. In Chapter 4, the results of the numerical solution of the mathematical model and its validation has been discussed. It also explores the potential situations to which the mathematical model can be applied. Finally, Chapter 5 highlights the main conclusions of the thesis and provides recommendations for future work.

CHAPTER 2

LITERATURE REVIEW

2.1 MATHEMATICAL MODELS OF PACKED BED SOLID STATE FERMENTATION BIOREACTORS

In the past decade, several mathematical models of packed bed solid state fermentation bioreactors have been developed. The important models are due to Saucedo-Castañeda et al.(1990), Sangsurasak and Mitchell (1995,1998),Hasan et al.(1998),Ashley et al.(1999),Mitchell et al.(1999) , Weber et al. (1999), Mitchell and Von Meien (2000) ,Mitchell et al. (2002),Weber et al. (2002) , Von Meien and Mitchell (2002) , Dos Santos et al. (2004) , Von Meien et al.(2004).

The various assumptions included by authors in the development of mathematical models is the visualization of the packed bed solid state fermentation bioreactor as pseudohomogeneous or a two phase system, prominence of conduction or convection in heat transfer, rate of biomass formation depending upon logistic growth kinetics or oxygen consumption.

Broadly the mathematical models for packed bed solid state fermentation bioreactors developed till date can be classified into two broad categories:-

- i) Mathematical models treating air and the substrate bed as a pseudohomogeneous phase.
- ii) Mathematical models treating air and the substrate bed as two distinct phases.

In the first category, the models developed do not consider the mass transfer of water. They are primarily concerned with temperature profiles.

The second category of mathematical models not only takes into account heat transfer effects but also the mass transfer of water between the two phases. With the help of these models, the significance of phenomena like the drying of the substrate bed is also emphasized.

2.2 MATHEMATICAL MODELS TREATING AIR AND SUBSTRATE BED AS A PSEUDOHOMOGENEOUS PHASE

2.2.1 Model by Saucedo-Castañeda et al.(1990)

Objectives:-

- i) To identify strategies of temperature control.
- ii) To demonstrate the use of dimensionless numbers in the scale up of large fermentors.

Species: – Aspergillus niger

Substrate: – Cassava Wet Meal

Height of Bioreactor = 35 cm.

Radius of Bioreactor = 6 cm.

Temperature of Substrate Bed = 35°C

Transversal Velocity = 3500 cm/h

Assumptions:-

- 1) Material balance is not considered due to low diffusion rates of biomass and glucose in solid media.
- 2) Axial dispersion is not considered since length to diameter of particle ratio is 50.
- 3) Variations in θ -direction are not there.
- 4) Radial gradients dominate.

Method of Solution:-

Orthogonal Collocation was used to discretize the spatial terms and the resulting set of ordinary differential equations were solved by Runge-Kutta Method.

Constitutive Relationships Used:-

- 1) Carbon Dioxide reaction rate was taken into consideration.
- 2) Logistic Growth Kinetics.
- 3) Specific growth rate is empirically related with temperature.

Conclusions:-

- 1) Peclet number estimations indicate that conduction plays a major role.
- 2) Peclet and Biot Numbers can be used as scale up criteria.
- 3) Maintenance energy and inoculum size play a lesser role in the process.

2.2.2 Model by Sangsurasak and Mitchell(1995)

Objectives:-

- i) Identification of the dominant heat transfer mechanism.
- ii) Accounting the role of death of microorganisms due to an increase in temperature.
- iii) Proposing strategies to minimize temperature rise.

Species: - Rhizopus oligosporus

Substrate: - Starchy substrate

Height of Bioreactor = 0.3 m

Temperature of Substrate Bed = 35°C

Superficial Velocity = 0.3 m/s

Height to Diameter Ratio = 1.5

Assumptions:-

- 1) Air moves in axial direction only.
- 2) The presence of a water jacket maintains the outside surface of substrate mass at constant temperature.
- 3) Bed voidage does not change during fermentation.

Method of Solution:-

The equations were first written in a non-dimensional form and then orthogonal collocation was applied to convert the resulting set of partial differential equations into a set of ordinary differential equations which were solved by Backward Euler Method using a GEAR Fortran Subroutine.

Constitutive Relationships Used:-

- 1) Specific growth rate is a quadratic function of temperature.
- 2) Death occurred due to first order kinetics and specific death follows an Arrhenius relationship.
- 3) Logistic Growth Kinetics.

Conclusions:-

- 1) Convection is the dominant heat transfer mechanism.
- 2) Radial temperature gradients can be neglected.
- 3) At height to diameter ratio of 1:1 and below the temperature profiles were less pronounced.

2.2.3 Model by Sangsurasak and Mitchell (1998)

Objectives:-

- i) Development of a two dimensional model which describes heat transfer in axial and radial directions.
- ii) Determination of sensitivity of the model to thermal and substrate properties.

Species: - Aspergillus niger

Substrate: - Cassava and Wheat Bran

Height and Radius of Bioreactors at which study is conducted:-

i) 0.345 m. and 0.075 m.; ii) 0.35 m. and 0.03 m.

Temperature of Substrate Bed =35°C

Superficial velocities at which study is conducted:-

i) 0.0047m/s,0.0141 m/s, 0.0235m/s; ii) 0.01m/s

Assumptions:-

- 1) Existence of thermal and moisture equilibrium between substrate bed and air.
- 2) Bed voidage does not change during the process.
- 3) Mass transfer of water is neglected since growth is limited by higher temperatures.
- 4) Air moves in axial direction moves with a constant velocity profile.

Method of Solution:-

Orthogonal Collocation, using Jacobi Polynomials was used to discretize the spatial coordinates as a two dimensional axi-symmetric problem, leaving a set of ordinary differential equations which were solved by using GEAR package.

Constitutive Relationships employed:-

- 1) Logistic Growth Kinetics.
- 2) Specific growth rate is empirically related to temperature.

Conclusions:-

- 1) The effect of bed voidage, when varied from 0.2 to 0.7 in the simulation had a significant effect.
- 2) Specific growth rate was varied from 0.1 to 0.5 h⁻¹ and was identified as a key kinetic parameter.
- 3) By varying the thermal conductivity of the substrate bed from 0.5 to 1 W/mK, noticeable changes were observed.

2.2.4 Model by Hasan et al. (1998)

Objectives:-

- i) Development of a two dimensional mathematical model to simulate the effects observed when operating conditions of aeration and bed porosity are varied.
- ii) To study the time profiles of reducing sugars, glucoamylase productivity.

Species: - *Aspergillus niger*

Substrate: - Rice bran

Height of Bioreactor = 0.3 m

Radius of Bioreactor = 2.35 cm.

Temperature of Substrate Bed = 26°C

Superficial Velocity = 10 m/hr.

Assumptions:-

- 1) The system is non-adiabatic and the process is non-isothermal.
- 2) The pseudohomogeneous phase is isotropic in nature.
- 3) Air flows in axial direction.

Method of Solution:-

The fully implicit finite difference method was used to convert the set of partial differential equations into a set of ordinary differential equations and numerical integration was carried out for small time steps.

Constitutive Relationships:-

- 1) The temperature dependence on growth was described by a maximum specific growth rate constant and maximum biomass concentration.
- 2) Thermal conductivity of rice bran was correlated with density and moisture.
- 3) Logistic Growth Kinetics
- 4) Equations for sugar consumption and CO₂ formation were employed.

Conclusions:-

- 1) The optimum conditions found by simulations found for a 300 mm. high and 47 mm. diameter bioreactor are 30°C and 60 ml/g h flow of air.
- 2) Forced aeration in Solid State Fermentation can be used for temperature control.

Observation:-

- 1) In the simulation for temperature at the centre of the bed, for different bed porosities, the time at which maxima is attained does not change in Sangsurasak and Mitchell (1998), but in this model it varies.

2.2.5 Model by Mitchell et al. (1999)

Objectives:-

- i) To identify scale up strategies for packed bed bioreactors.

Species: - *Aspergillus niger*

Substrate: - Moist starchy substrate (Wheat Bran)

Height of Bioreactor = 1 m

Temperature of Substrate Bed = 30°C

Superficial Velocity = 0.01 m/s to 0.1 m/s

Assumptions:-

- 1) Heat transfer in axial direction is considered. In a large diameter bed, radial heat transfer is assumed to be negligible.
- 2) Air and moist solid at any particular location within the bed are assumed to be in thermal equilibrium.
- 3) Maintenance metabolism, effects of microbial growth on particle size and pressure drop across the bed are ignored.
- 4) Void fraction does not change with time.

Method of Solution:-

Orthogonal collocation using Jacobi Polynomials was used to discretize the spatial coordinate, leaving a set of ordinary differential equations which were then solved by GEAR package.

Constitutive Relationships used:-

- 1) Logistic Growth Kinetics
- 2) Specific growth rate is a function of temperature.

Conclusions:-

- 1) A modified Damköhler Number was proposed which was shown to be more useful than the Peclet and Biot number approach proposed by Saucedo-Castañeda et al. (1990).
- 2) The Damköhler number approach was found to be less useful than the scale up procedures based on dynamic heat transfer model. Damköhler number could be used by practitioners who do not have the resources to solve partial differential equations.

2.2.6 Model by Ashley et al. (1999)

Objective:-

- i) To employ axial heat transfer model to identify strategies for prevention of temperatures reaching undesirable levels.

Species: - *Aspergillus niger*

Substrate: - Moist starchy substrate

Height of Bioreactor = 0.345 m

Temperature of Substrate Bed = 30°C

Superficial Velocity = 0.0236 m/s

Assumptions:-

- 1) The bed is assumed to be wide enough so that heat transfer in the horizontal (radial) direction is negligible.
- 2) Maintenance metabolism is ignored.
- 3) Effect of microbial growth on particle size and pressure drop are not considered.
- 4) Void fraction does not change with time.

Method of Solution:-

Orthogonal collocation was used to discretize the axial coordinate, resulting in a set of ordinary differential equations which were solved by GEAR. The model of the well mixed bed with which comparisons were made was solved by using Fourth-Order Runge Kutta Method.

Constitutive Relationships:-

- 1) Specific growth rate is a function of temperature.
- 2) Logistic Growth Kinetics.

Conclusions:-

- 1) Periodic Air Reversal was found to be a less useful temperature control strategy.
- 2) If the species can tolerate mixing events of the frequency of 10 to 60 per hour, then periodic mixing can be very useful in preventing overheating in packed bed solid state fermentation bioreactors.

2.2.7 Model of Mitchell and Von Meien(2000)

Objective:-

- i) To provide guidelines for optimum design and operation for Zymotis Bioreactors (Packed Bed Bioreactors with internal cooling plates)

Species: - *Aspergillus niger*

Substrate: - Whole cassava meal

Height of Bioreactor = 0.3 m

Temperature of Substrate Bed = 35°C

Superficial Velocity = 0.01 m/s

Assumptions:-

- 1) In the vertical direction, convection dominates. Hence conduction in vertical direction is neglected.
- 2) Air flows in the vertical direction; hence there is no horizontal convection.
- 3) The reported dividing and mixing of air leads to eddy conduction. It is assumed to be smaller than static conduction.
- 4) Void fraction does not change with time.
- 5) Air and moist solid at a particular location are assumed to be in thermal equilibrium.

Method of Solution:-

The method of characteristics is adopted to the equation system which was converted to dimensionless form. The modified system was solved by applying polynomial approximation using zeroes of Jacobi Polynomials as interpolation points. The resulting differential-algebraic system was solved by using DASSL routine.

Constitutive Relationships Employed:-

- 1) Logistic Growth Kinetics.
- 2) Specific growth rate is a function of temperature.
- 3) Density and thermal conductivity were evaluated as volume-weighted averages, whereas specific heat was evaluated as a mass weighted average.

Conclusions:-

- 1) The best strategy to operate a Zymotis Packed Bed Bioreactor was found to be the use of a small spacing between cooling plates, high superficial velocity and to vary the cooling water temperature.

2.2.8 Model by Mitchell et al. (2002)

Objective:-

- i) To analyze the performance of a Zymotis Bioreactor by investigating the optimal value for the spacing between its cooling plates.

Species: - Aspergillus niger

Substrate: - Starchy substrate

Height = 2.5 m

Superficial Air Velocity = 0.01m/s

Plate half spacing = 0.03 m

Assumptions:-

- 1) Air flows in the vertical direction, hence vertical conduction is ignored.
- 2) Void fraction does not change with time.
- 3) The system is modelled such that the heat transfer in centre is responsible for removing heat from the two half slabs of fermenting substrate.

Constitutive Relationships:-

- 1) Logistic Growth Kinetics
- 2) Density and Thermal Conductivity are calculated as volume weighted averages, whereas heat capacity is determined as a mass-weighted average of substrate bed and air properties.

Method of Solution:-

The system of model equations was non-dimensionalized and solved by method of characteristics using Orthogonal Collocation applied in both horizontal and vertical dimensions. The resulting differential algebraic system was solved by DASSL routine.

- C
1)
2)
- 3) In the accumulation terms of all balances, contribution of gases and all mass accumulation terms are negligible.
 - 4) Pseudosteady state with respect to temperature and oxygen consumption is assumed.
 - 5) Axial gradients in oxygen consumption are neglected.
 - 6) Bed porosity is constant.

2.

Constitutive Relationships:-

O
i)

- 1) Biomass production rate, water and substrate consumption rates are all related to oxygen production rates.

Conclusions:-

S_f
S_i

- 1) In this paper, a model has been developed in which a water balance has been incorporated to predict the water content of the solid substrate in a packed bed.
- 2) Oxygen consumption rate is used to estimate heat production rate which also includes the heat generated by maintenance metabolism.

H
D
T
S

2.3.2 Model of Weber et al. (2002)

A
1)

Objective:-

- i) To validate the model proposed in 1999 (2.3.1) with experiments in a 15 dm³ packed bed bioreactor.

2)

Species: - *Coniothyrium minitans*

Substrate: - Hemp and Oats

3)

Diameter of the bioreactor = 20 cm.

Height of the bioreactor = 70 cm. (Bed Height = 50 cm.)

Superficial aeration rate = 0.027 – 0.069 kg/m³s

C

1

2

Assumptions:-

- 1) The water present in the biomass and in the substrate is not discriminated.
- 2) Air is in equilibrium with the solid matrix at any point in the bed.

- 3) Pseudosteady state with respect to temperature and oxygen consumption rate.
- 4) In the accumulation term of all balances, the contribution of gases and all mass accumulation terms are negligible.

Constitutive Relationships Used:-

- 1) Logistic Growth Kinetics, where in Rathowsky equation is employed for describing temperature dependency on maximum specific growth rate and maintenance requirements.
- 2) Oxygen consumption rate is described by linear growth model.

Computational Methods Used:-

Three Dimensional Regression was done on Sigma Plot 4.0.1 to evaluate unknown parameters in logistic law, linear growth model and Rathowsky equation.

Conclusions:-

- 1) The proper choice of a solid substrate is essential since drying of the solid substrate can occur.
- 2) Shrinkage of Solid Substrate and channelling can have severe effects on the process.

2.3.3 Model of Von Meien and Mitchell (2002)

Objective:-

- i) To develop a two phase dynamic model that describes heat and mass transfer in intermittently mixed solid state fermentation bioreactors.

Species: - *Aspergillus niger*

Substrate: - Corn grits

Inlet air flow rate = $0.06 \text{ kg/m}^2 \text{ s}$

Cross-sectional area of the bioreactor = 1 m^2

Height of the bioreactor = 2.5 m

Assumptions:-

- 1) The bioreactor is assumed to be wide enough so that heat transfer to walls makes a negligible contribution to cooling.
- 2) The microorganisms can tolerate infrequent mixing events.
- 3) Growth is completely inhibited during the mixing event. At the end of the mixing event, solid and gas phase temperatures are assumed to have returned to inlet air temperature.

Constitutive Relationships:-

- 1) Logistic Growth Kinetics.
- 2) Isotherm for corn grits.
- 3) Specific growth rate is evaluated on the basis of specific growth rate based on water activity and on temperature.

Method of Solution:-

In the system of partial differential equations developed, the spatial derivative was approximated by finite differences using equal size elements, which was solved by DASSL routine.

Conclusions:-

- 1) The model emphasizes the loss of water content in the solid which may occur due to evaporation of water in the substrate.

2.3.4 Model of Von Meien et al. (2004)

Objective:-

- i) To test different control strategies based on classical proportional-integral – derivative and advanced dynamic matrix control algorithm for an intermittently-mixed, forcefully aerated solid state fermentation bioreactor.

Species: - *Aspergillus niger*

Substrate: - Corn grits

Assumptions and Constitutive Relationships:-

The model equations and constitutive relationships are the same as Von Meien and Mitchell (2002).

Method of Solution:-

The same method of solution has been used as by Von Meien and Mitchell (2002).

Conclusions:-

- 1) Significant temperature gradients are unavoidable in large solid state fermentation bioreactors.
- 2) An easier strategy which could be implemented is the control of bed temperature and moisture content by manipulating air at 100% humidity. The intermittent mixing event could be triggered by a drop in humidity of outlet to replenish water.

2.4 SIGNIFICANCE OF MATHEMATICAL MODELS

The significance of the mathematical models developed till date are given in the following table .The table aims to highlight the key features associated with the aforesaid mathematical models of Packed Bed Solid State Fermentation Bioreactors. It also reflects upon the method of computation used to solve the model.

Table 2-1: Significance of Mathematical Models for Packed Bed Solid State Fermentation Bioreactors

Author	Significance	Method of Solution
Saucedo-Castañeda et al.(1990)	Quantitative relationships regarding growth of <i>Aspergillus niger</i> were established from experiments.	Orthogonal Collocation ,Runge Kutta Method
Sangsurasak and Mitchell (1995)	Demonstration by two-dimensional mathematical model that higher temperatures attained in Packed Bed Solid State Fermentation Bioreactors can cause significant death.	Orthogonal Collocation , Backward Euler Method
Sangsurasak and Mitchell (1998)	Bed porosity and specific growth rate were established as significant parameters with the help of a two dimensional mathematical model.	Orthogonal Collocation
Hasan et al. (1998)	Experiments and simulations conducted for <i>Aspergillus niger</i> on rice bran.	Fully implicit finite differences

Ashley et al. (1999)	Overheating in packed bed solid state fermentation bioreactors could be reduced by periodic mixing.	Orthogonal Collocation
Mitchell et al. (1999)	The Mathematical Modeling approach could be employed as a scaleup tool and it was proved to be better than the Peclet and Biot number approach as proposed by Saucedo-Castaneda et al.(1990).	Orthogonal Collocation
Weber et al.(1999)	Inclusion of a water mass balance to the model equations. They emphasized the threat which drying of substrate bed could cause in Solid State Fermentation bioprocesses.	
Mitchell and Von Meien (2000)	Development of a mathematical model of Zymotis Bioreactor.	The model equations were solved by polynomial approximation.

Mitchell et al.(2002)	A method to evaluate the plate spacing between heat transfer plates in Zymotis packed bed bioreactors is proposed.	Orthogonal Collocation
Von Meien and Mitchell(2002)	Development of a two-phase mathematical model which incorporates solution of energy and mass balance equations for air and substrate bed.	Finite Difference Method
Weber et al.(2002)	Experimental validation of the model equations proposed in 1999 for Coniothryium minitans.	
Dos Santos et al.(2004)	Propose the potential threat caused to enzyme production by thermal denaturation in packed beds.	Runge-Kutta Method (Well Mixed Bed)
Von Meien et al.(2004)	Implementation of various control strategies to intermittently mixed packed bed bioreactors	Finite Difference Method.

	lead to the conclusion that significant temperature gradients are unavoidable in Solid State Fermentation Bioreactors.	
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2.5 MOTIVATION FOR THE PRESENT STUDY:

From the literature review, it is clearly evident that significant temperature gradients cannot be avoided in Packed Bed Solid State Fermentation Bioreactors. The mathematical models developed for packed beds till date involve partial differential equations which have been solved in most studies using Orthogonal Collocation.

It is hence pertinent to search for methods which would facilitate the design of Packed Bed Solid State Fermentation Bioreactors. Our work has two primary aims:-

- i) To reduce the complexities involved in analysis and computation.
- ii) To obtain the time profiles of various characterizing variables like temperature, substrate bed water content at different heights ,so that it could aid in determining the optimum dimensions of the packed bed bioreactor.

The industrial application of Dos Santos et al. (2004) assumes vital significance as it emphasizes the threat which the presence of temperature gradients poses to production of enzymes.

Till date, the most comprehensive model developed for Packed Bed Solid State Fermentation Bioreactor is that of Von Meien and Mitchell (2002) which explains the underlying heat and mass transfer phenomena in both the phases (substrate bed and air).

Hence the aim of this study would be to explore new insights and further avenues for research employing approaches in Chemical Reaction Engineering with the information incorporated in the aforesaid models.

2.6 CONCLUDING REMARKS

In this chapter ,the mathematical model describing the heat and mass transfer phenomena in packed bed solid state fermentation bioreactors by various research workers have been discussed with respect to their objectives ,assumptions ,physical dimensions ,constitutive relationships ,solution methodologies and pertinent conclusions. An evaluation of mathematical models citing their significance was done to derive conclusions regarding future scope of work

CHAPTER 3

DEVELOPMENT OF THE MATHEMATICAL MODEL FOR A PACKED BED SOLID STATE FERMENTATION BIOREACTOR USING N-TANKS IN SERIES APPROACH

3.0 INTRODUCTION

In this chapter, mathematical models have been developed for packed bed solid state fermentation bioreactors using the N- Tanks in Series approach. Two cases have been discussed .A packed bed reactor can be designed akin to a plug flow reactor. The performance of equal sized mixed flow reactors in series (when $N \rightarrow \infty$) approaches to plug flow conditions (Levenspiel, 1999).The primary advantage of applying the equal sized mixed flow reactors in series approach to the packed bed solid state fermentation bioreactor is that the solution of a partial differential equation is simplified to the solution of a set of simultaneous ordinary differential equations.

3.1 MATHEMATICAL MODEL FOR A PSEUDO HOMOGENEOUS PHASE

The first mathematical model developed using the approach is for the production of protease by *Penicillium fellutanum* as studied by Dos Santos et al., 2004; where a 1m. high and 1 m diameter packed bed bioreactor has been modelled as a single well mixed continuous stirred tank reactor.

The packed bed bioreactor in our model has been modelled as a plug flow reactor which is visualized as equal sized mixed tank bioreactors

connected in series .The model equations in this section have been developed for two mixed flow bioreactors connected in series, which has been later generalized for the number of tanks, n.

3.1.1 Assumptions

The assumptions employed in the derivation of the mathematical model are:-

1. The specific growth rate is assumed to be unaffected by changes in temperature.
2. The specific heat is assumed independent of temperature.
3. No pressure drop occurs when air flows from one mixed tank to the other.
4. Moisture and thermal equilibrium exists between air and the solids in the bed.
5. The kinetic energy, potential energy and the shaft work terms have been neglected.
6. The reference temperature for the calculation of enthalpy has been taken as 0°C.
7. The flow rate of cooling water in the water jacket is assumed to be high enough so that no temperature change occurs in it.
8. The water content of the bed in all the visualized mixed tanks is the same.
9. The void fraction does not change with time.
10. The effect of particle size and pressure drop has not been considered.

3.1.2 Derivation of the Mathematical Model

The mathematical model is derived for two mixed tanks connected in series .The mixed tanks have equal cross sectional areas, heights and heat transfer areas for water jackets.

In this section, the model equations have been developed simultaneously for both the bioreactors. The air to the first bioreactor is supplied at the provided inlet conditions. The air at the inlet to the second bioreactor is at the condition, which prevails at the exit of the first reactor.

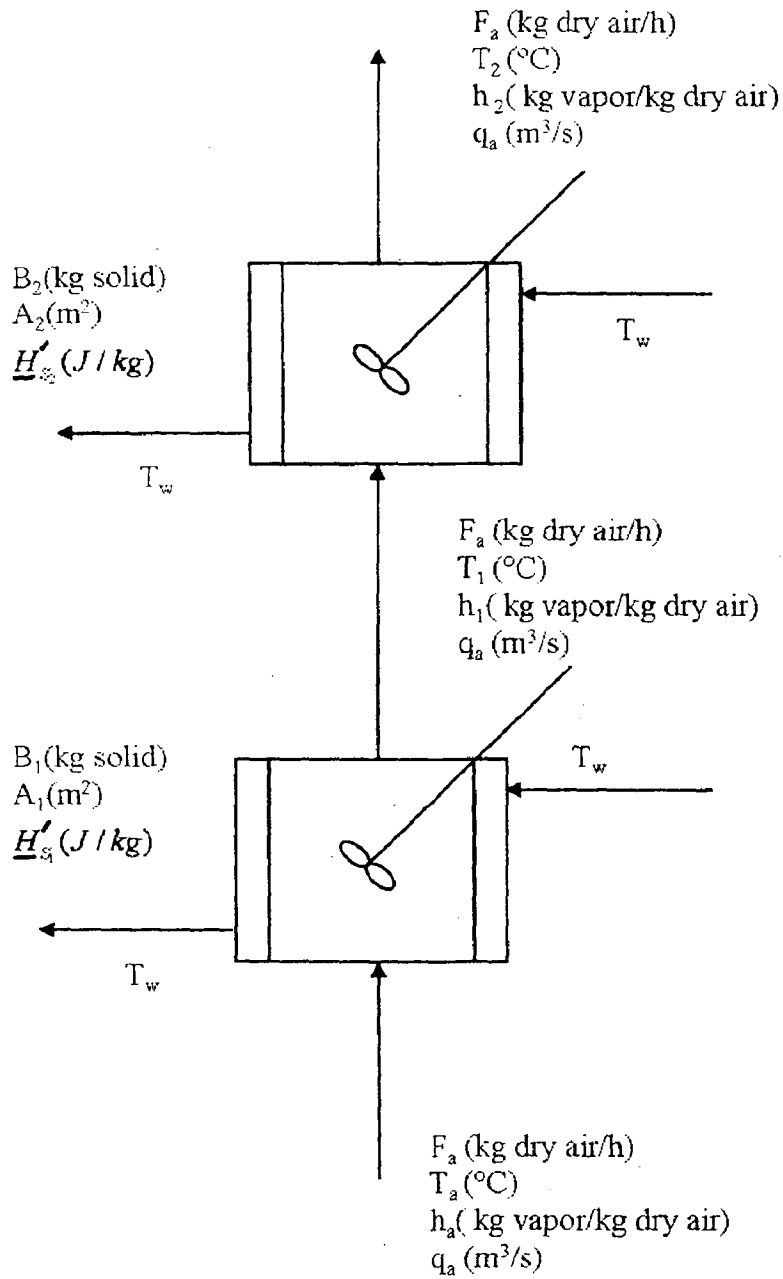


Figure 3.1 A schematic representing the system of bioreactors to be modeled as a pseudohomogeneous phase system

Energy Balance on the system yields:-

Bioreactor 1:-

$$\frac{d}{dt}(U_1 + KE_1 + PE_1) = q_a \rho_a (\underline{U}_a + \underline{KE}_a + \underline{PE}_a) - q_a \rho_a (\underline{U}_{e_1} + \underline{KE}_{e_1} + \underline{PE}_{e_1}) + Q_1 + W_{T_1}$$

(3.0(a))

Bioreactor 2:-

$$\frac{d}{dt}(U_2 + KE_2 + PE_2) = q_a \rho_a (\underline{U}_{e_1} + \underline{KE}_{e_1} + \underline{PE}_{e_1}) - q_a \rho_a (\underline{U}_{e_2} + \underline{KE}_{e_2} + \underline{PE}_{e_2}) + Q_2 + W_{T_2}$$

(3.0(b))

The total work in the system consists of work done upon the system by virtue of flow and shaft work.

i.e.

$$W_{T_1} = q_a P_{f_1} - q_a P_{e_1} + W_{sh}$$

(3.1(a))

$$W_{T_2} = q_a P_{f_2} - q_a P_{e_2} + W_{sh}$$

(3.1(b))

The enthalpy of the system is defined as:

$$H'_s = U + PV$$

(3.2)

$$\frac{dH'_s}{dt} = \frac{dU}{dt} + \frac{d(PV)}{dt}$$

(3.3)

For a fluid-solid system

$$\frac{d(PV)}{dt} = 0$$

$$\therefore \frac{dU}{dt} = \frac{dH'_s}{dt} \quad (3.4)$$

Substituting the relationships 3.1 and 3.4 in the corresponding equations

Bioreactor 1:-

$$\frac{dH'_{S_1}}{dt} = q_a \rho_a \underline{U}_a - q_a \rho_a \underline{U}_{e_1} + Q_1 + q_a P_{f_1} - q_a P_{e_1} \quad (3.5(a))$$

$$\frac{dH'_{S_1}}{dt} = q_a \rho_a \left[\underline{U}_a + \frac{P_{f_1}}{\rho_a} \right] - q_a \rho_a \left[\underline{U}_{e_1} + \frac{P_{e_1}}{\rho_a} \right] + Q_1$$

Bioreactor 2:-

$$\frac{dH'_{S_2}}{dt} = q_a \rho_a \underline{U}_{e_1} - q_a \rho_a \underline{U}_{e_2} + Q_2 + q_a P_{e_1} - q_a P_{e_2} \quad (3.5(b))$$

$$\therefore (P_{f_2} = P_{e_1})$$

$$\frac{dH'_{S_2}}{dt} = q_a \rho_a \left[\underline{U}_{e_1} + \frac{P_{e_1}}{\rho_a} \right] - q_a \rho_a \left[\underline{U}_{e_2} + \frac{P_{e_2}}{\rho_a} \right] + Q_2$$

The enthalpy per unit mass is defined as:

$$\underline{H}'_s = \underline{U} + \frac{P}{\rho} \quad (3.6)$$

$$F = q\rho \quad (3.7)$$

Substituting (3.6) and (3.7) in (3.5), the following expressions are obtained

Bioreactor 1:-

$$\frac{dH'_{S_1}}{dt} = F_a \underline{H}'_{S_a} - F_a \underline{H}'_{S_1} + Q_1 \quad (3.8(a))$$

Bioreactor 2:-

$$\frac{dH'_{S_2}}{dt} = F_a \underline{H}'_{S_1} - F_a \underline{H}'_{S_2} + Q_2 \quad (3.8(b))$$

The enthalpy of the system is defined as:-

$$H'_S = mC_p dT \quad (3.9)$$

For a solid state fermentation bioreactor unit, enthalpy is defined as:

$$H'_{S_1} = B_1(1+W)C_{pb}dT \quad (3.10(a))$$

$$H'_{S_2} = B_2(1+W)C_{pb}dT \quad (3.10(b))$$

The specific enthalpy of air entering the bioreactor can be defined as

$$\underline{H}'_{S}(T) = C_{pa}(T - T_{ref}) + h_a |_T [C_{pv}(T - T_{ref}) + \lambda] \quad (3.11)$$

At $T_{ref} = 0^\circ\text{C}$

$$\underline{H}'_{S}(T) = C_{pa}T + h_a |_T [C_{pv}T + \lambda] \quad (3.12)$$

The heat transfer term Q , accounts for two mechanisms:-

i) Microorganisms evolving heat in the control volume

$$= (\text{Mass of solids})(\text{Heat yield due to growth})(\text{Rate of biomass formation})$$

$$= BY_Q \frac{dX}{dt} \quad (3.13)$$

ii) Heat transferred to the water circulating in the jacket

$$= -hA(T - T_w) \quad (3.14)$$

Therefore

$$Q = BY_Q \frac{dX}{dt} - hA(T - T_w) \quad (3.15)$$

If the system is modeled as two continuous stirred tank bioreactor in series system, then

$$B_1 = B_2 = B/2 \quad (3.16)$$

$$A_1 = A_2 = A/2 \quad (3.17)$$

Hence employing the relations, 3.10, 3.12, and 3.15-3.17 in 3.8 the following expressions are obtained as:

Bioreactor 1:-

$$\frac{dH'_{S_1}}{dt} = F_a H'_{S_a} - F_a H'_{S_1} + Q_1 \quad (3.8(a))$$

$$B_1(1+W)C_{ph} \frac{dT_1}{dt} = F_a [C_{pa}T_a + h_a |_{T_a} (C_{pv}T_a + \lambda)] - F_a [C_{pa}T_1 + h_a |_{T_1} (C_{pv}T_1 + \lambda)] \\ + B_1 Y_Q \frac{dX}{dt} - hA_1(T_1 - T_w) \quad (3.18(a))$$

Bioreactor 2:-

$$\frac{dH'_{S_2}}{dt} = F_a H'_{S_1} - F_a H'_{S_2} + Q_2 \quad (3.8(b))$$

$$B_2(1+W)C_{ph} \frac{dT_2}{dt} = F_a [C_{pa}T_1 + h_a |_{T_1} (C_{pv}T_1 + \lambda)] - F_a [C_{pa}T_2 + h_a |_{T_2} (C_{pv}T_2 + \lambda)] \\ + B_2 Y_Q \frac{dX}{dt} - hA_2(T_2 - T_w) \quad (3.18(b))$$

The relations 3.18 can be written as:

Bioreactor 1:-

$$\frac{dT_1}{dt} = \frac{\left[F_a C_{pa} (T_a - T_1) + F_a \lambda (h_a |_{T_a} - h_a |_{T_1}) + F_a C_{pv} (h_a |_{T_a} T_a - h_a |_{T_1} T_1) + \left(\frac{B}{2} \right) Y_Q \frac{dX}{dt} - h \left(\frac{A}{2} \right) (T_1 - T_w) \right]}{\left(\frac{B}{2} \right) (1+W) C_{ph}} \quad (3.19(a))$$

Bioreactor 2:-

$$\frac{dT_2}{dt} = \frac{\left[F_a C_{pa} (T_1 - T_2) + F_a \lambda (h_a |_{T_1} - h_a |_{T_2}) + F_a C_{pv} (h_a |_{T_1} T_1 - h_a |_{T_2} T_2) + \left(\frac{B}{2} \right) Y_Q \frac{dX}{dt} - h \left(\frac{A}{2} \right) (T_2 - T_w) \right]}{\left(\frac{B}{2} \right) (1+W) C_{pb}}$$

(3.19(b))

For the nth Bioreactor, the above equation may be generalized as:

$$\frac{dT_n}{dt} = \frac{\left[F_a C_{pa} (T_{n-1} - T_n) + F_a \lambda (h_a |_{T_{n-1}} - h_a |_{T_n}) + F_a C_{pv} (h_a |_{T_{n-1}} T_{n-1} - h_a |_{T_n} T_n) + \left(\frac{B}{n} \right) Y_Q \frac{dX}{dt} - h \left(\frac{A}{n} \right) (T_n - T_w) \right]}{\left(\frac{B}{n} \right) (1+W) C_{pb}}$$

(3.20)

3.1.3 Constitutive Relationships and Parameters Employed

3.1.3.1 Growth Kinetics

Logistic growth kinetics was assumed in terms of biomass content per kg of initial dry substrate (IDS).

$$\frac{dX}{dt} = \mu X \left(1 - \frac{X}{X_m} \right)$$

(3.21)

The above relationship stresses the concept that solid state fermentation is a contact process limited by available surface area with a maximal biomass packing density related with X_m and not included in the conventional Monod Model. It also incorporates the assumption of μ being independent of substrate concentration (Saucedo-Castañeda et al., 1990).

3.1.3.2. Enzyme Activity and Denaturation

The experimental protease activity profile is described by-

$$\frac{dE}{dt} = \alpha(E + D_e) \left(1 - \frac{(E + D_e)}{E_m} \right) - kE \quad (3.22)$$

The denaturation rate constant is described by the function

$$k = A_0 \exp \left(\frac{-En_d}{R(T + 273.15)} \right) \quad (3.23)$$

The denatured enzyme is produced by a first order denaturation reaction

$$\frac{dD_e}{dt} = kE \quad (3.24)$$

3.1.3.3 Evaluation of Humidity

The humidity at the desired temperature was evaluated by

$$P_{vap} = \exp \left(18.3036 - \frac{3816.44}{(T + 273.15) - 46.13} \right) \quad (3.25)$$

$$h_u = 0.6246 \left(\frac{P_{vap}}{P - P_{vap}} \right) \quad (3.26)$$

The pressures in the above correlation are to be used in mmHg and temperatures in °C.

3.1.3.4 Geometric Considerations

The packed bed bioreactor is of cylindrical shape; hence the quantity of dry solids in the bioreactor is given by

$$B = \rho_b L \pi \left(\frac{D}{2} \right)^2 \quad (3.27)$$

The area of heat transfer is given by

$$A = \pi DL \quad (3.28)$$

3.1.3.5 Temperature Control Strategy

The following equation adjusts the temperature of cooling water in response to the bed temperature.

$$T_w = T_s - F_g(T - T_s) \quad (3.29)$$

3.1.3.6 Parameters

The following values of the parameters were employed in the solution of the model equations. (Dos Santos et al., 2004).

Table-3-1: Various parameters and their value employed for the solution of model based on pseudohomogeneous phase system

Parameter	Value
A_0	$3.447 \times 10^{59} \text{h}^{-1}$
C_{pa}	$1007 \text{ J kg dry air}^{-1} \text{ } ^\circ\text{C}^{-1}$
C_{pb}	$1867 \text{ J kg solid}^{-1} \text{ } ^\circ\text{C}^{-1}$
C_{pv}	$2500 \text{ J kg vapor}^{-1} \text{ } ^\circ\text{C}^{-1}$
E_{na}	$364070 \text{ J mol}^{-1}$
E_m	141.3 PU/g IDS
F_g	0 or 2 (as indicated)
h	$360000 \text{ J h}^{-1} \text{ m}^{-2} \text{ } ^\circ\text{C}^{-1}$
P	760 mm Hg
R	$8.314 \text{ J mol}^{-1} \text{ } ^\circ\text{C}^{-1}$
W	1 kg water/kg IDS
X_m	0.250 kg biomass/kg IDS
Y_Q	$8.366 \times 10^6 \text{ J/kg biomass}$
α	0.0782 h^{-1} (Penicillium fellutanum) 0.2 h^{-1} (Aspergillus niger)
λ	$2500900 \text{ J kg vapor}^{-1}$

μ	0.0782 h ⁻¹ (<i>Penicillium fellutanum</i>) 0.2 h ⁻¹ (<i>Aspergillus niger</i>)
ρ_b	178.5 kg dry solids /m ³

3.1.4 Summary of Model Equations to be solved

The following equations for the nth bioreactor have been developed and are to be solved simultaneously

$$\frac{dX_n}{dt} = \mu X_n \left(1 - \frac{X_n}{X_m} \right) \quad (3.30)$$

$$\frac{dT_n}{dt} = \frac{\left[F_a C_{pa} (T_{n-1} - T_n) + F_a \lambda (h_a |_{T_{n-1}} - h_a |_{T_n}) + F_a C_{pv} (h_a |_{T_{n-1}} T_{n-1} - h_a |_{T_n} T_n) + \left(\frac{B}{n} \right) Y_Q \frac{dX}{dt} - h \left(\frac{A}{n} \right) (T_n - T_w) \right]}{\left(\frac{B}{n} \right) (1+W) C_{pb}} \quad (3.31)$$

$$\frac{dD_{e_n}}{dt} = kE_n \quad (3.32)$$

$$\frac{dE_n}{dt} = \alpha (E_n + D_{e_n}) \left(1 - \frac{(E + D_{e_n})}{E_m} \right) - kE_n \quad (3.33)$$

3.1.5 Initial Conditions

The initial conditions for solving all the above equations are as follows:-

At t=0, in all bioreactors

i) Initial biomass content of the solid substrate is
 $X_0 = 0.002 \text{ kg biomass/kg IDS} \quad (3.34)$

ii) Temperature of all the n visualized well mixed bioreactors
 $T = 30^\circ\text{C} \quad (3.35)$

iii) The denatured enzyme in all stages is

$$D_{e_i} = 0 \text{ PU/g IDS} \quad (3.36)$$

iv) Solid substrate active enzyme content is

$$E_0 = 1 \text{ PU/g IDS} \quad (3.37)$$

The model equations were solved for a 1m diameter and 1 m height bioreactor as had been done by Dos Santos et al.(2004).The air flow rate , F_a was 60 kg dry air/hr.

3.1.6 Method of Solution and Computer Program

To solve the set of simultaneous ordinary differential equations, (3.30-3.33) employing the mixed tanks-in series approach, a computer program in in MATLAB 6.1. (Math Works Inc.) was developed.

The numerical method used for the solution is Runge-Kutta-Fehlberg method (Matthews and Fink, 2004) without step size control (Appendix A).

The main inputs to the computer program are:-

1. Flow rate of air entering the bioreactor.
2. Temperature of the inlet air.
3. Solid Substrate Active Enzyme Content.
4. Number of continuous mixed flow tank reactors in which the packed bed bioreactor is visualized to be divided.
5. Denatured Enzyme Content.

The main outputs of the computer program are the profiles of the following variables with time for the visualized continuous stirred tank reactors.

1. Temperature of the bioreactor
2. Biomass production
3. Protease Enzyme Activity
4. Denaturation of Protease Enzyme

The complete computer program may be obtained from the author or from his supervisor on request.

3.2 MATHEMATICAL MODEL FOR A TWO PHASE SYSTEM

The second mathematical model which has been developed using the N CSTR's in series approach considers air(gaseous) and the dry solid bed as two distinct phases as compared to the earlier model where the two phases – gaseous and solid were modelled as a pseudohomogeneous phase.

Von Meien and Mitchell (2002) considered the growth of *Aspergillus niger* on corn wherein a non-equilibrium situation was described between the gas and solid phases. They have considered mass transfer of water between the two phases. Only Weber et al. (1999, 2002) had incorporated a water balance in his model and thus considered the resulting mass transfer effects.

3.2.1 Derivation of the Mathematical Model

The mathematical model has been derived for a cylindrical shaped packed bed reactor. The equations for two equal sized mixed tank in series system has been developed which later has been generalized for n mixed tanks in series.

3.2.2 Assumptions

The assumptions employed in the derivation of the mathematical model are:-

1. The bioreactor is assumed to be wide enough so that heat transfer to the walls makes a negligible contribution to cooling .Hence heat transfer in axial direction is considered.
2. The isotherm for corn grits has been assumed to be similar to that of the substrate.
3. The dry solid concentration and the biomass concentration do not vary in the bioreactor .

$$\text{i.e. } S_1 = S_2 = S_3 = \dots S_n = S$$

$$b_1 = b_2 = b_3 = \dots b_n = b$$

4. The reference temperature in the evaluation of convection terms(in the energy balances) is 0°C.
5. The void fraction does not change with time.
6. Effect of particle size and pressure drop has not been considered.
7. The gas flow rate per unit area does not change from bioreactor to bioreactor.

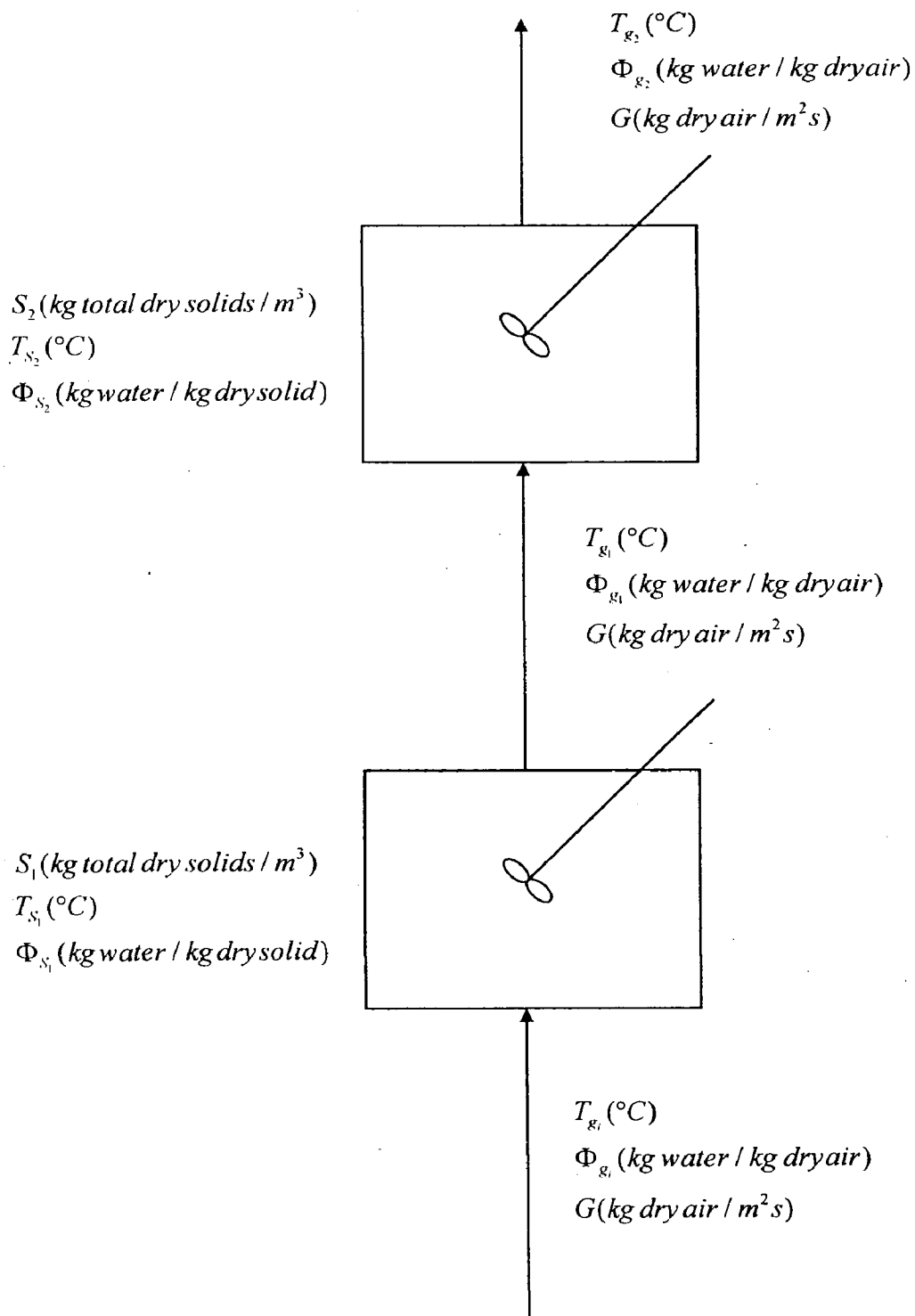


Figure 3.2 A schematic representing the system of bioreactors to be modeled as a two phase system

Hence,

$$\left(\begin{array}{c} \text{Rate of} \\ \text{accumulation of} \\ \text{water in solid phase} \\ \text{of bioreactor 1} \end{array} \right) = \left(\begin{array}{c} \text{Rate of water} \\ \text{generated by} \\ \text{biomass} \end{array} \right) - \left(\begin{array}{c} \text{Rate of water} \\ \text{transferred from} \\ \text{solids to air} \end{array} \right)$$

$$\frac{1}{\Delta t} \left[AH_1 \Delta (\Phi_{S_1} S_1) \right] = Y_{WB} \left[S_1 \frac{\Delta b}{\Delta t} + b \frac{\Delta S_1}{\Delta t} \right] AH_1 - K_a (\Phi_{S_1} - \Phi_{S_1}^*) AH_1 \quad (3.47(a))$$

Divide by AH_1 and taking $\text{Lt } \Delta t \rightarrow 0$

$$\frac{d(\Phi_{S_1} S_1)}{dt} = Y_{WB} \left[S_1 \frac{db}{dt} + b \frac{dS_1}{dt} \right] - K_a (\Phi_{S_1} - \Phi_{S_1}^*) \quad (3.48(a))$$

$$\Phi_{S_1} \frac{dS_1}{dt} + S_1 \frac{d\Phi_{S_1}}{dt} = Y_{WB} \left[S_1 \frac{db}{dt} + b \frac{dS_1}{dt} \right] - K_a (\Phi_{S_1} - \Phi_{S_1}^*) \quad (3.49(a))$$

$$\frac{d\Phi_{S_1}}{dt} = \frac{1}{S_1} \left[Y_{WB} \left(S_1 \frac{db}{dt} + b \frac{dS_1}{dt} \right) - K_a (\Phi_{S_1} - \Phi_{S_1}^*) - \Phi_{S_1} \frac{dS_1}{dt} \right] \quad (3.50(a))$$

Bioreactor 2:-

Rate of accumulation of water in the solid phase

$$= \frac{1}{\Delta t} \left[AH_2 \Delta (\Phi_{S_2} S_2) \right] \quad (3.44(b))$$

Rate of transfer of water from solids to gas through evaporation

$$= K_a (\Phi_{S_2} - \Phi_{S_2}^*) AH_2 \quad (3.45(b))$$

$$\text{Rate of water generated from biomass} = Y_{WB} \left[\frac{\Delta(bS_2)}{\Delta t} \right] AH_2$$

$$= Y_{WB} \left[S_2 \frac{\Delta b}{\Delta t} + b \frac{\Delta S_2}{\Delta t} \right] AH_2 \quad (3.46(b))$$

Hence,

$$\left(\begin{array}{c} \text{Rate of} \\ \text{accumulation of} \\ \text{water in solid phase} \\ \text{of bioreactor 2} \end{array} \right) = \left(\begin{array}{c} \text{Rate of water} \\ \text{generated by} \\ \text{biomass} \end{array} \right) - \left(\begin{array}{c} \text{Rate of water} \\ \text{transferred from} \\ \text{solids to air} \end{array} \right)$$

$$\frac{1}{\Delta t} \left[AH_2 \Delta (\Phi_{S_2} S_2) \right] = Y_{WB} \left[S_2 \frac{\Delta b}{\Delta t} + b \frac{\Delta S_2}{\Delta t} \right] AH_2 - K_a (\Phi_{S_2} - \Phi_{S_2}^*) AH_2 \quad (3.47(b))$$

Divide by AH_2 and taking $Lt \Delta t \rightarrow 0$

$$\frac{d(\Phi_{S_2} S_2)}{dt} = Y_{WB} \left[S_2 \frac{db}{dt} + b \frac{dS_2}{dt} \right] - K_a (\Phi_{S_2} - \Phi_{S_2}^*) \quad (3.48(b))$$

$$\Phi_{S_2} \frac{dS_2}{dt} + S_2 \frac{d\Phi_{S_2}}{dt} = Y_{WB} \left[S_2 \frac{db}{dt} + b \frac{dS_2}{dt} \right] - K_a (\Phi_{S_2} - \Phi_{S_2}^*) \quad (3.49(b))$$

$$\frac{d\Phi_{S_2}}{dt} = \frac{1}{S_2} \left[Y_{WB} \left(S_2 \frac{db}{dt} + b \frac{dS_2}{dt} \right) - K_a (\Phi_{S_2} - \Phi_{S_2}^*) - \Phi_{S_2} \frac{dS_2}{dt} \right] \quad (3.50(b))$$

3.2.3.3 Equations for the production of biomass

For bioreactors 1 and 2

$$\frac{db}{dt} = \mu b \left(1 - \frac{b}{b_m} \right) \quad (3.51)$$

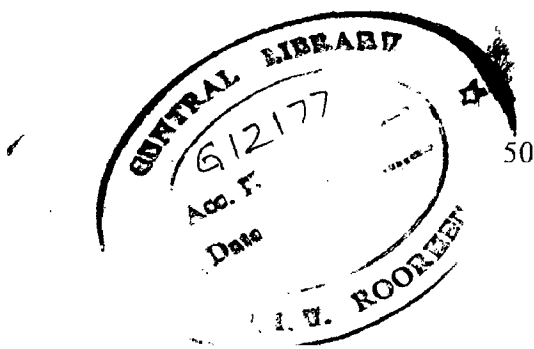
3.2.3.4 Material Balance on Dry Solids

Bioreactor 1:-

$$\frac{dS_1}{dt} = Y_{SB} \frac{d(bS_1)}{dt} \quad (3.52(a))$$

$$\frac{dS_1}{dt} = Y_{SB} \left(b \frac{dS_1}{dt} + S_1 \frac{db}{dt} \right) \quad (3.53(a))$$

$$\frac{dS_1}{dt} = \frac{Y_{SB} S_1}{(1 - b Y_{SB})} \frac{db}{dt} \quad (3.54(a))$$



Bioreactor 2:-

Rate of energy accumulation in the substrate bed

$$= \frac{1}{\Delta t} S_2 (C_{pv} + \Phi_{S_2} C_{pw}) \Delta T_{S_2} (AH_2) \quad (3.61(b))$$

Rate of energy transferred due to evaporation of water from the substrate bed

$$= \lambda K_a (\Phi_{S_2} - \Phi_{S_2}^*) AH_2 \quad (3.62(b))$$

Rate of energy transferred from gas to substrate bed=

$$= h_r AH_2 (T_{g_2} - T_{S_2}) \quad (3.63(b))$$

Rate of energy generated by biomass production

$$= Y_Q \frac{\Delta(bS_2)}{\Delta t} AH_2 \quad (3.64(b))$$

Formulation of energy balance on the substrate bed yields:-

$$\left(\begin{array}{c} \text{Rate of energy} \\ \text{accumulated in} \\ \text{the solid phase in} \\ \text{Bioreactor 2} \end{array} \right) = \left(\begin{array}{c} \text{Rate of} \\ \text{energy} \\ \text{transferred} \\ \text{from gas to} \\ \text{solid phase} \end{array} \right) - \left(\begin{array}{c} \text{Rate of energy} \\ \text{transferred due} \\ \text{to evaporation} \\ \text{of water from} \\ \text{solid phase} \end{array} \right) + \left(\begin{array}{c} \text{Rate of} \\ \text{energy} \\ \text{generated} \\ \text{by biomass} \\ \text{production} \end{array} \right)$$

$$\frac{1}{\Delta t} S_2 (C_{pv} + \Phi_{S_2} C_{pw}) \Delta T_{S_2} (AH_2) = h_r AH_2 (T_{g_2} - T_{S_2}) - \lambda K_a (\Phi_{S_2} - \Phi_{S_2}^*) AH_2 + Y_Q \frac{\Delta(bS_2)}{\Delta t} AH_2$$

(3.65(b))

Dividing all the terms by AH_2 and taking the $Lt \Delta t \rightarrow 0$

$$S_2(C_{ps} + \Phi_{S_2} C_{pw}) \frac{dT_{S_2}}{dt} = h_t(T_{g_2} - T_{S_2}) - \lambda K_a(\Phi_{S_2} - \Phi_{S_2}^*) + Y_{O_2} \frac{d(bS_2)}{dt} \quad (3.66(b))$$

$$S_2(C_{ps} + \Phi_{S_2} C_{pw}) \frac{dT_{S_2}}{dt} = h_t(T_{g_2} - T_{S_2}) - \lambda K_a(\Phi_{S_2} - \Phi_{S_2}^*) + Y_{O_2} \left(S_2 \frac{db}{dt} + b \frac{dS_2}{dt} \right) \quad (3.67(b))$$

3.2.4 Constitutive Relationships

3.2.4.1 Initial Water Content for Air

The moisture content for air entering into the packed bed bioreactor is at an initial water activity of 0.99.

$$P_w^{sat} = 133.322 \exp \left(18.3036 - \frac{3816.44}{(T_g + 273.15) - 46.13} \right) \quad (3.68)$$

At $T_g = 35^\circ\text{C}$ (Temperature of air at inlet)

$$P_w^{sat} = 5601.93 \text{ Pa}$$

$$P = 101325 \text{ Pa}$$

The water activity of the entering gas is evaluated as:

$$a_{wg} = \frac{\Phi_g P}{P_w^{sat} (\Phi_g + 0.62413)} \quad (3.69)$$

$$0.99 = \frac{\Phi_g (101325)}{5601.93 (\Phi_g + 0.62413)}$$

$$\Phi_g = 0.03614 \text{ kg water/kg dry air}$$

3.2.4.2 Initial Water Content for Solids

The moisture content of the substrate bed has an initial water activity of 0.99. The initial water activity of the solid phase is evaluated as:-

$$a_{w,x} = \left\{ 1 - \exp\left[-\Phi_s^{(c3+c4T_s)} \cdot \exp(c1 + c2T_s)\right] \right\}^{\frac{1}{c5}} \quad (3.70)$$

The constants for evaluation are:-

$$c1 = 2.9$$

$$c2 = 0.004$$

$$c3 = 1.275$$

$$c4 = -0.0029$$

$$c5 = 0.32$$

$$(0.99)^{0.32} = 1 - \exp\left[-\Phi_s^{(1.275+(-0.0029 \times 35))} \exp(2.9 + (0.004)(35))\right]$$

$$\Phi_s = 0.33245 \text{ kg water/kg dry solid}$$

3.2.4.3 Mass Transfer Coefficient

The coefficient for mass transfer of water between the gas phase and substrate bed is evaluated by the following correlation.

$$K_a = \left(a1 + a2 \left[T_g + 273 \right] \right) \Phi_s - a3 + a4(T_g + 273) \quad (3.71)$$

3.2.4.4 Heat Transfer Coefficient

The coefficient for convective heat transfer between the substrate bed and gas phase is evaluated by the following correlation developed for drying of corn.

$$h_i = 44209.85 \left[\frac{G(T_g + 273)}{0.0075P} \right]^{0.6011} \quad (3.72)$$

3.2.4.5 Specific Growth Rate

Temperature and the substrate bed water activity affect the specific growth rate. The relative specific growth rates are defined on the maximum specific growth rate at optimal conditions. The fractional specific growth rate based μ_w based on water activity is given by:-

$$\mu_w = \exp(d1.a_{w,x}^3 + d2.a_{w,x}^2 + d3.a_{w,x} + d4) \quad (3.73)$$

The fractional specific growth rate μ_T is calculated based on temperature employing

$$\mu_T = \frac{1}{\mu_{opt}} \frac{A_T \exp\left(\frac{-E_{A1}}{R(T_S + 273)}\right)}{1 + B_T \exp\left(\frac{-E_{A2}}{R(T_S + 273)}\right)} \quad (3.74)$$

The actual specific growth rate is evaluated as the geometric mean of the two specific growth rates.

$$\mu = \mu_{opt} \sqrt{\mu_W \mu_T} \quad (3.75)$$

3.2.5 Model Equations for the nth bioreactor:

3.2.5.1 Mass Balance for water in gas phase

$$\varepsilon \rho_g \frac{d\Phi_{g_n}}{dt} = G \left[\frac{\Phi_{g_{n-1}} - \Phi_{g_n}}{(H/n)} \right] + K_a (\Phi_{s_n} - \Phi_{s_n}^*) \quad (3.76)$$

3.2.5.2 Mass Balance for water in the substrate bed

$$\frac{d\Phi_{s_n}}{dt} = \frac{1}{S} \left[Y_{WB} \left(S \frac{db}{dt} + b \frac{dS}{dt} \right) - K_a (\Phi_{s_n} - \Phi_{s_n}^*) - \Phi_{s_n} \frac{dS}{dt} \right] \quad (3.77)$$

3.2.5.3 Energy Balance for the gas phase

$$\varepsilon \rho_g \left(C_{pg} + \Phi_{g_n} C_{pv} \frac{dT_{g_n}}{dt} \right) = G \left(C_{pg} + \Phi_{g_n} C_{pv} \right) \left[\frac{T_{g_{n-1}} - T_{g_n}}{(H/n)} \right] - h_1 (T_{g_n} - T_{s_n}) \quad (3.78)$$

3.2.5.4 Energy Balance for the solid phase

$$S (C_{ps} + \Phi_{s_n} C_{pv}) \frac{dT_{s_n}}{dt} = h_1 (T_{g_n} - T_{s_n}) - \lambda K_a (\Phi_{s_n} - \Phi_{s_n}^*) + Y_Q \left(S \frac{db}{dt} + b \frac{dS}{dt} \right) \quad (3.79)$$

3.2.5.5 Equation for balance for dry solids

$$\frac{dS}{dt} = \frac{Y_{SB} S}{(1 - b Y_{SB})} \frac{db}{dt} \quad (3.80)$$

3.2.5.6 Equation for biomass production

$$\frac{db}{dt} = \mu b \left(1 - \frac{b}{b_m} \right) \quad (3.81)$$

3.2.6 Parameters for the solution of Mathematical Model

Table-3-2: Various parameters and their values employed for the solution of model based on two- phase system

Parameter	Value employed in Solution
a1	7.304
a2	1.77×10^{-2}
a3	2.202
a4	-6.18×10^{-3}
A_T	$7.483 \times 10^7 \text{ s}^{-1}$
b_m	0.250 kg biomass/kg total dry solids
B_T	$1.300 \times 10^{47} \text{ s}^{-1}$
c1	2.9
c2	0.004
c3	1.275
c4	-0.0029
c5	0.32
C_{pg}	1005 J/kg dry air °C
C_{ps}	2500 J/kg dry solid °C
C_{pv}	1791 J/kg water vapor °C
C_{pw}	4184 J/kg water °C
d1	618.9218
d2	-1863.527
d3	1865.097
d4	-620.6684

E_{A1}	70225 J/mol
E_{A2}	283356 J/mol
P	101325 Pa
R	8.314 J/mol °C
Y_{SB}	-2 kg total dry solids/kg biomass
Y_Q	8.366×10^6 J/kg biomass
Y_{WB}	0.3 kg water/kg biomass
ϵ	0.35 m^3 void space/ m^3
λ	2414300 J/kg water
ρ_g	1.14 kg dry air/ m^3
ρ_s	300 kg substrate/ m^3 moist substrate
μ_{opt}	$9.00535 \times 10^{-5} \text{ s}^{-1}$

3.2.7 Initial Conditions

The initial conditions for solving the above equations are as follows:

At $t=0$, in all bioreactors

i) Initial biomass content of the solid substrate bed

$$X_0 = 0.002 \text{ kg biomass/kg solid} \quad (3.82)$$

ii) Temperature of the inlet air

$$T_g = 35 \text{ °C} \quad (3.83)$$

iii) Temperature of the substrate bed

$$T_s = 35 \text{ °C} \quad (3.84)$$

iv) Initial moisture content of the substrate bed

$$\Phi_s = 0.33245 \text{ kg water/ kg dry solid} \quad (3.85)$$

v) Moisture content of the entering dry air

$$\Phi_g = 0.03614 \text{ kg water/kg dry air} \quad (3.86)$$

vi) Substrate Concentration (kg/m^3)

$$S = (1 - \epsilon)\rho_s$$

$$S = (1 - 0.35) \times 300 = 195 \text{ kg}/\text{m}^3 \quad (3.87)$$

The model equations for solved for a 2.5 m high bioreactor as done by Von Meien and Mitchell (2002).

3.2.8 Method of Solution and Computer Program

To solve the set of simultaneous ordinary differential equations (3.76-3.81) employing the mixed tanks in series approach, a computer program in MATLAB 6.1. (Math Works Inc.) was developed.

The numerical method used for the solution is the Fourth Order Runge Kutta Method (Matthews and Fink , 2004) without step size control.(Appendix B)

The main inputs to the computer program are:-

1. Flow rate of air entering in the bioreactor.
2. Temperature of the inlet air.
3. Temperature of the water bed.
4. Water content of air entering the bioreactor.
5. Water content of the substrate bed.
6. Initial biomass content in the substrate bed.
7. Height of the bioreactor.
8. Number of continuous mixed flow reactors in which the Packed Bed Solid State Fermentation Bioreactors is visualized to be divided.

The main outputs of the computer program are the profiles of the following variables with time for the visualized continuous stirred tank reactors.

1. Temperature of the solid bed.
2. Temperature of the air.
3. Biomass concentration.
4. Solid phase Concentration.
5. Water content of the air.
6. Water content of the substrate bed.

The complete computer program may be obtained from the author or from his supervisor on request.

3.3 CONCLUDING REMARKS

In this chapter, two mathematical models for Packed Bed Solid State Fermentation Bioreactor have been developed, using the N-Tanks in Series approach. The first one treats the solids and the air as a pseudohomogeneous phase, and the other treats them as distinct phases with mass transfer of water occurring between them. The constitutive relationships necessary for evaluation and the values of parameters required to solve the mathematical model are also stated.

RESULTS AND DISCUSSION

4.0 INTRODUCTION

The mathematical model of a Packed Bed Solid State Fermentation Bioreactor employing the N-Tanks in Series approach envisaging two cases has been developed. The first mathematical model (3.1) was developed using the pseudohomogeneous approach where the packed bed has the properties which are the mass weighted averages of air (gaseous phase) and the substrate (solid phase).

The second mathematical model (3.2) considers that the substrate and air as two distinct phases of the packed bed. In this chapter, the numerical solution of the model equations has been presented and discussed.

4.1 RESULTS FROM THE PSEUDOHOMOGENEOUS MODEL

The aim of the pseudohomogeneous model is to determine the temperature, solid substrate active enzyme content and denatured enzyme profiles with respect to time for a slower growing organism (*Penicillium fellutanum*) and for a faster growing organism (*Aspergillus niger*). The effect of the gain factor in the temperature control strategy and the variation of the aforesaid variables with height was also found out. The results of the pseudohomogeneous model are validated with the experimental results of Ghildyal et al. (1994) determined for *Aspergillus niger*, and a comparison with the two-dimensional model (solved by applying Orthogonal Collocation in radial and axial directions) of Sangsurasak and Mitchell (1998) is also made. The solid substrate active enzyme content and the denatured enzyme levels have been defined in terms of protease units, where Dos Santos et al. (2004) has defined it as the amount of enzyme that produces a difference in A_{440} of 1.0 absorbance unit per minute in the assay.

4.1.1 Profiles of Temperature , Solid Substrate Active Enzyme Content and Denatured Enzyme for Aspergillus niger

The determination of temperature, solid substrate active enzyme content and denatured enzyme was carried out by varying the number of tanks, N. All the other parameters were kept same. The profiles for N=1(Fig. 4.1 - 4.4), N=5(Figs. 4.9,4.11,4.13) and for N=10 (Fig. 4.21-4.24). The following were observed in regard to the aforesaid:-

1. The temperature profiles for N=1(Fig. 4.1) indicates a maximum temperature of 39.6°C at a time of 24 hours. On observing the profiles for temperature for N=5(Fig. 4.9), the maximum temperature is 41.4°C, and for N=10(Fig. 4.21), the maximum temperature attained at 24 hours is 41.6°C. It clearly indicates that a bed of 1m height cannot be designed as a single well mixed bed. Instead the N-Tanks of series approach would not only represent the situation clearly, but also provide the temperature profiles at intermediate heights.
2. The profile of Solid Substrate Active Enzyme Content for a single well mixed bed (Fig. 4.2) represents that there is no active enzyme content after 30 hours in the whole bed. On the other hand, the N-Tanks in Series Model reveals that about 50% of the bed has an active enzyme content above 10 PU/g IDS(Fig. 4.22).
3. The denatured enzyme –time profiles for N=1 (Fig. 4.3) indicates a peak denaturation of 132.6 PU /g IDS. The denaturation profile for N=5(Fig. 4.13) and for N=10 (Fig. 4.23) indicate a peak denaturation of 140 PU / g IDS and a denaturation of above 120 PU /g IDS for 50% of the bed.
4. The biomass profiles for N=1 (Fig. 4.4) and N=10(Fig. 4.24) are identical since a constant specific growth rate has been assumed.

4.1.2 Profiles of Temperature , Solid Substrate Active Enzyme Content and Denatured Enzyme for Penicillium fellutanum

The determination of temperature, solid substrate active enzyme content and denatured enzyme was carried out by varying the number of tanks, N. All the other parameters were kept same. The profiles for N=1(Fig. 4.5 - 4.8),N=5(Figs. 4.15,4.17,4.19) and for N=10 (Fig. 4.25-4.28).The following were observed in regard to the aforesaid:-

1. The temperature profiles for N=1(Fig. 4.5) indicate a maximum temperature of 34.6°C at a time of 62 hours .On observing the profiles for temperature for N=5(Fig. 4.15) and for N=10(Fig. 4.25) is 34.6°C.The identical temperature obtained in the case of Penicillium fellutanum is attributed to its slower growing characteristics, which in turn reemphasizes the importance of N-Tanks in Series Model for the prediction of faster growing biological species like Aspergillus niger.
2. The profile of Solid Substrate Active Enzyme Content for a single well mixed bed (Fig. 4.6) represents that the active enzyme content at the end of 140 hours is 62 PU/g IDS. The N-Tanks in Series Model wherein reveals that about 50% of the bed has an active enzyme content above 60 PU/g IDS (Fig. 4.26).
3. The denatured enzyme –time profiles for N=1 (Fig. 4.7) indicates a peak denaturation of 80 PU /g IDS. The denaturation profile for N=5(Fig. 4.19) and for N=10 (Fig. 4.27) indicate a peak denaturation of 93 PU / g IDS and a denaturation of above 80 PU /g IDS for 50% of the bed. The results indicate that the whole bed does not attain a denaturation enzyme level of 80 PU/g IDS as indicated by the results of a single mixed bed.
4. The biomass profiles for N=1 (Fig. 4.8)and N=10(Fig. 4.28) are identical since a constant specific growth rate has been assumed .

4.1.3 Effect of the gain factor in temperature control strategy

The effect of the gain factor F_g for controlling the cooling water temperature was studied for Aspergillus niger(Fig. 4.9-4.14) and Penicillium

fellutanum (Fig. 4.15-4.20).The findings for *Aspergillus niger* are summarized as below:-

- i) The peak temperature at $t=24$ hours decreased from 41.4°C to 34.3°C when the gain factor was increased from 0 to 2 (Fig. 4.9 -4.10).
- ii) If the gain factor is increased by 2, then the solid substrate active enzyme content never falls to zero. Instead the enzyme content is always above 97 PU/g IDS (Fig. 4.11-4.12).
- iii)The peak denaturation level falls drastically from 142 PU/g IDS to 45 PU/g IDS.(Fig. 4.13-4.14)

In the case of *Penicillium fellutanum*, the following were observed:-

- i) The peak temperature at $t=62$ hours decreased from 34.6°C to 31.7°C when the gain factor was increased from 0 to 2 (Fig. 4.15 -4.16).
- ii) If the gain factor is increased by 2, then the solid substrate active enzyme content never falls to zero. Instead the enzyme content is always above 97 PU/g IDS (Fig. 4.17-4.18).
- iii)The peak denaturation level falls drastically from 142 PU/g IDS to 45 PU/g IDS.(Fig. 4.19-4.20).

From the above observations it can be concluded that denaturation of enzyme caused due to the presence of temperature gradients in packed bed solid state fermentation bioreactors could be prevented by varying the cooling temperature. Dos Santos et al. (2004) also elucidated the fact, but now it is possible to visualize and determine the enzyme content and denaturation levels at various heights with the help of N-Tanks in Series Model.

4.1.4 Study of the variation of Temperature, Solid Substrate Active Enzyme Content and Denatured Enzyme with height (axial distance)

The importance of the variation of temperature, solid substrate active enzyme content and denatured enzyme with height (axial distance) is that, it can help to provide the crucial design estimate regarding the probable height of the bioreactor in a simplified manner. It has been made possible only by the use of N-Tanks in Series Model.

For *Aspergillus niger* the Temperature (Fig. 4.29), Solid State Active Enzyme Content (Fig. 4.30) and Denatured Enzyme (Fig. 4.31) with distance are shown. It can be seen that at an axial distance of 0.4 m. the peak temperature at 24 hour is 38°C, the solid substrate active enzyme content at 25 hr. is 80 PU/g IDS and the enzyme denaturation level is of the order of 70 PU/g IDS. On the other hand at a height of 1m the peak temperature is 41.4°C, by 25 hrs. there is virtually no active enzyme left and denaturation level is of the order of 140 PU/g IDS. Hence it gives insight in the development of bioprocesses regarding what should be the probable height of the bioreactor and what enzyme denaturation levels can be tolerated.

In the case of *Penicillium fellutanum* insights of a similar nature can be obtained where the profiles of temperature (Fig. 4.32), Solid Substrate Active Enzyme (Fig. 4.33) and Denatured Enzyme (Fig. 4.34) are shown.

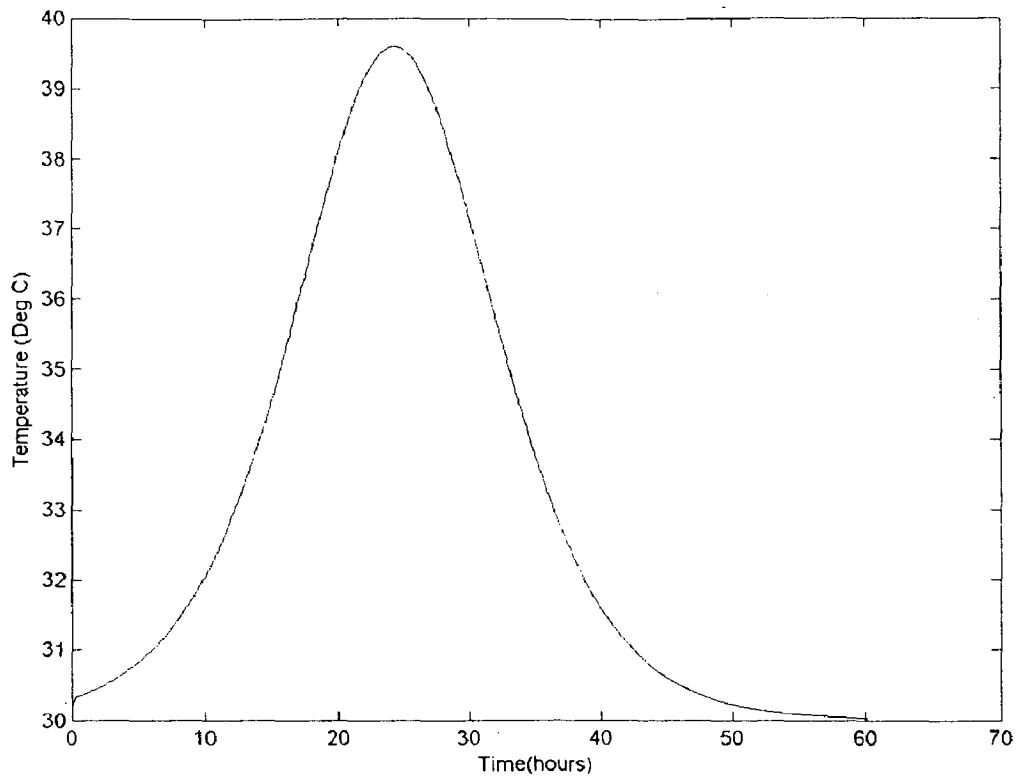


Figure 4.1 Plot of temperature vs. time for *Aspergillus niger* ($N=1, F_g=0, \mu=0.2, \alpha=0.2$)

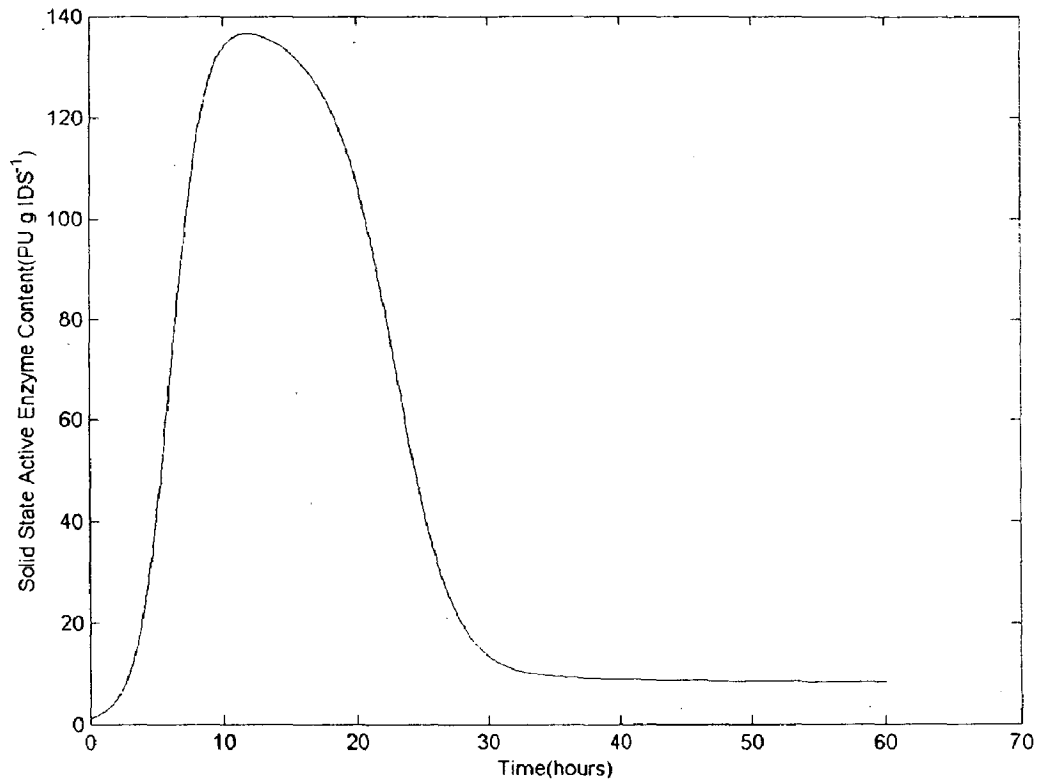
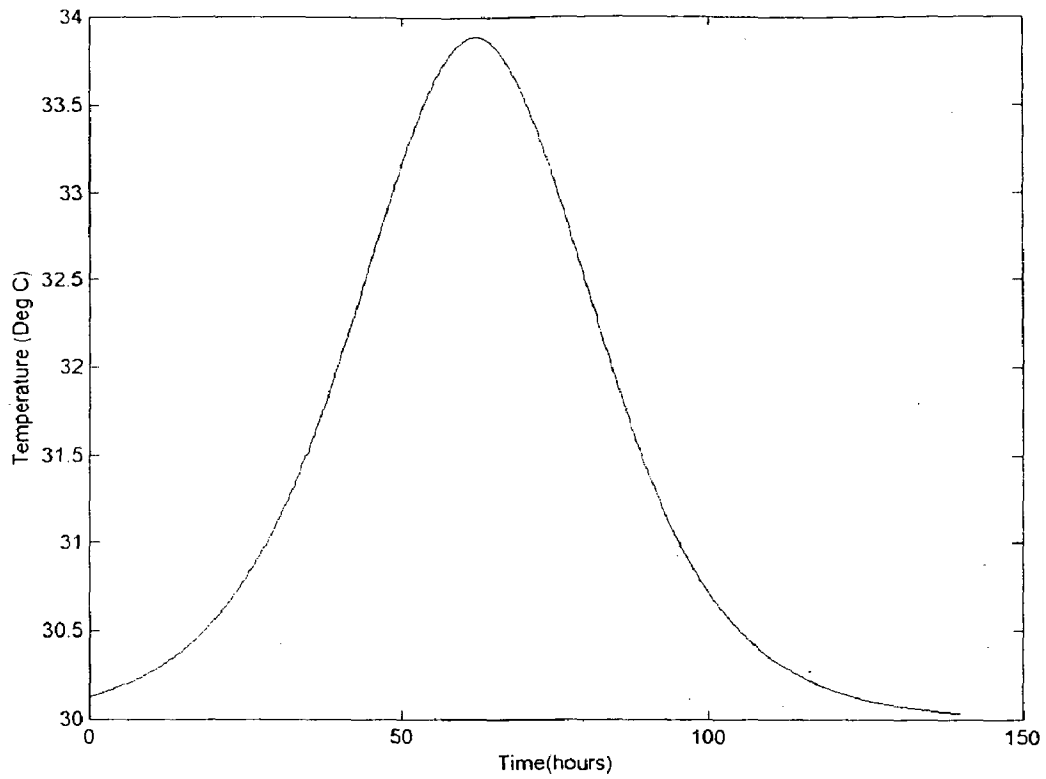
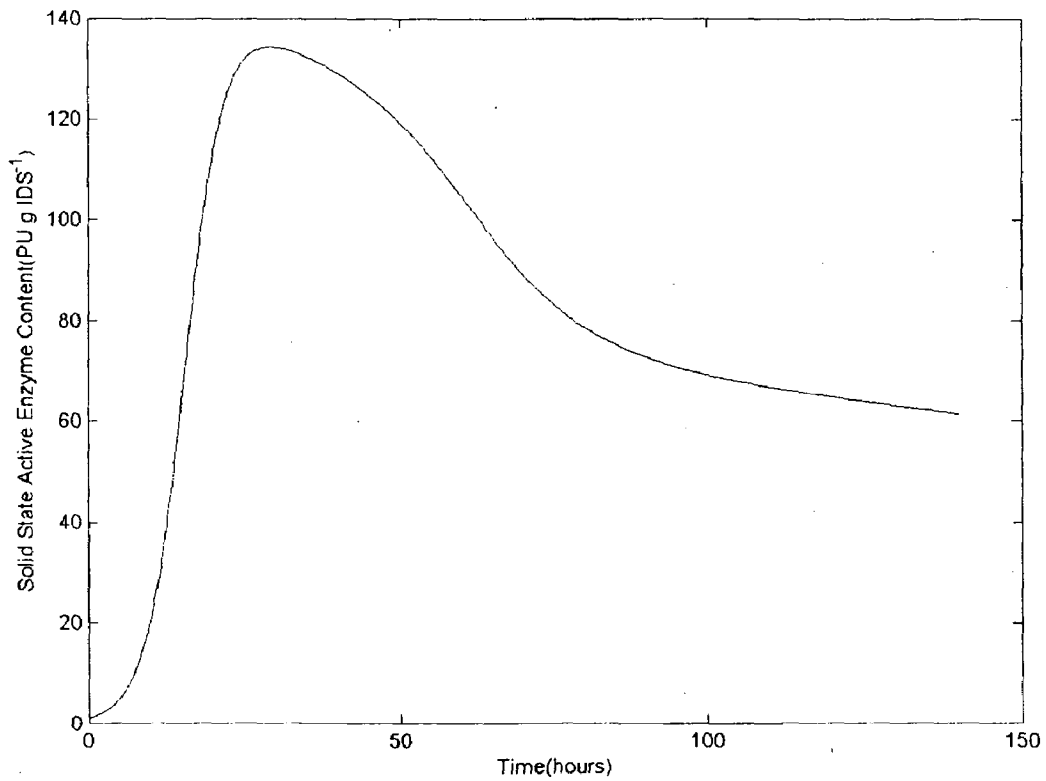


Figure 4.2 Plot of Solid State Active Enzyme Content vs. time for *Aspergillus niger* ($N=1, F_g=0, \mu=0.2, \alpha=0.2$).



**Figure 4.5 Plot of temperature vs. time for *Penicillium fellutanum*
($N=1, F_g=0, \mu=0.0782, \alpha=0.0782$)**



**Figure 4.6 Plot of Solid State Active Enzyme Content vs. time for *Penicillium fellutanum*
($N=1, F_g=0, \mu=0.0782, \alpha=0.0782$)**

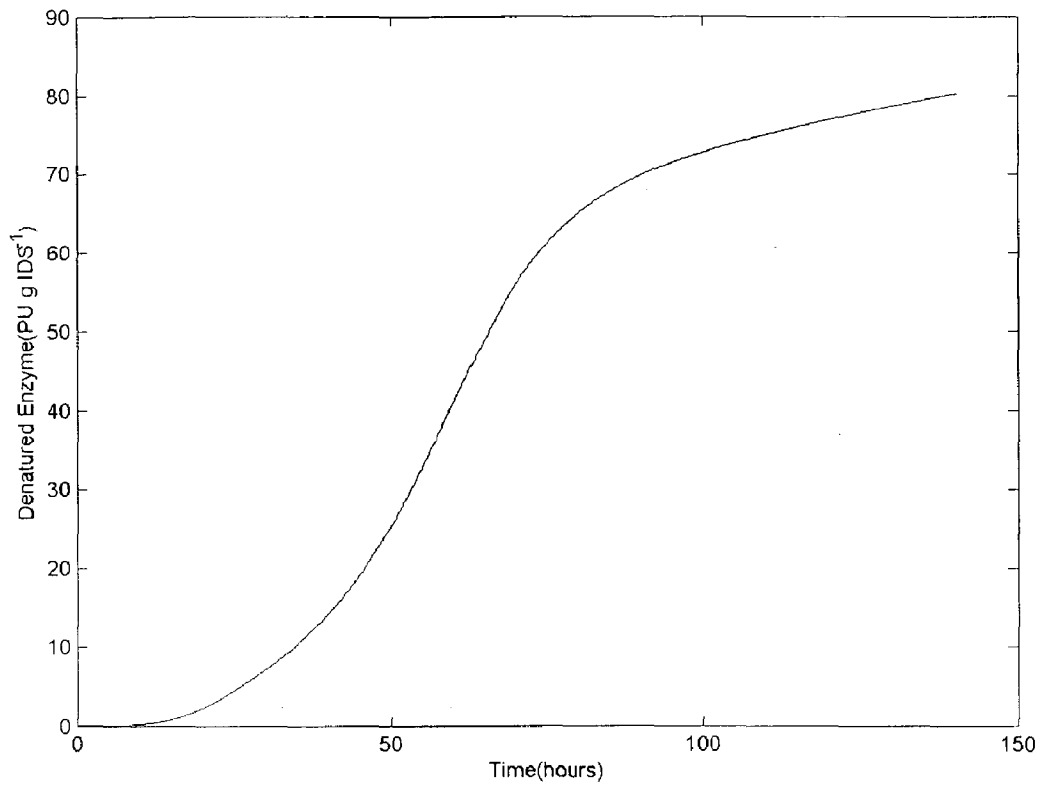


Figure 4.7 Plot of Denatured Enzyme vs. time for *Penicillium fellutanum*
 ($N=1, F_g=0, \mu=0.0782, \alpha=0.0782$)

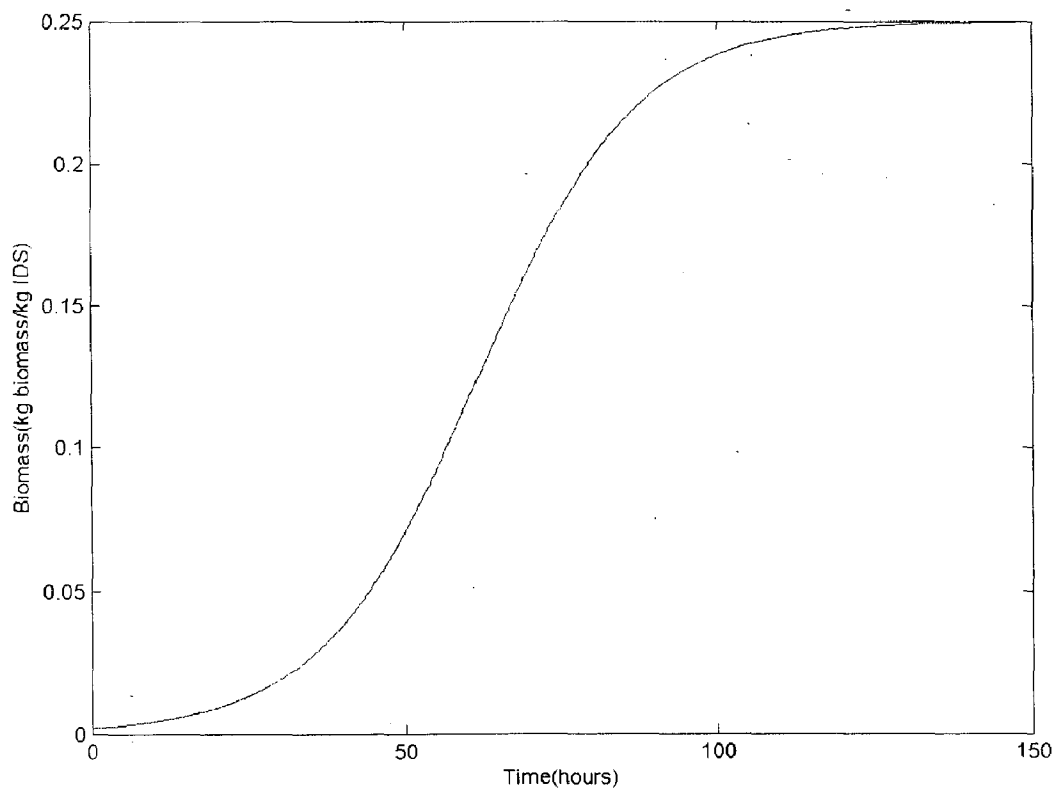


Figure 4.8 Plot of Biomass Content vs. time for *Penicillium fellutanum*
 ($N=1, F_g=0, \mu=0.0782, \alpha=0.0782$)

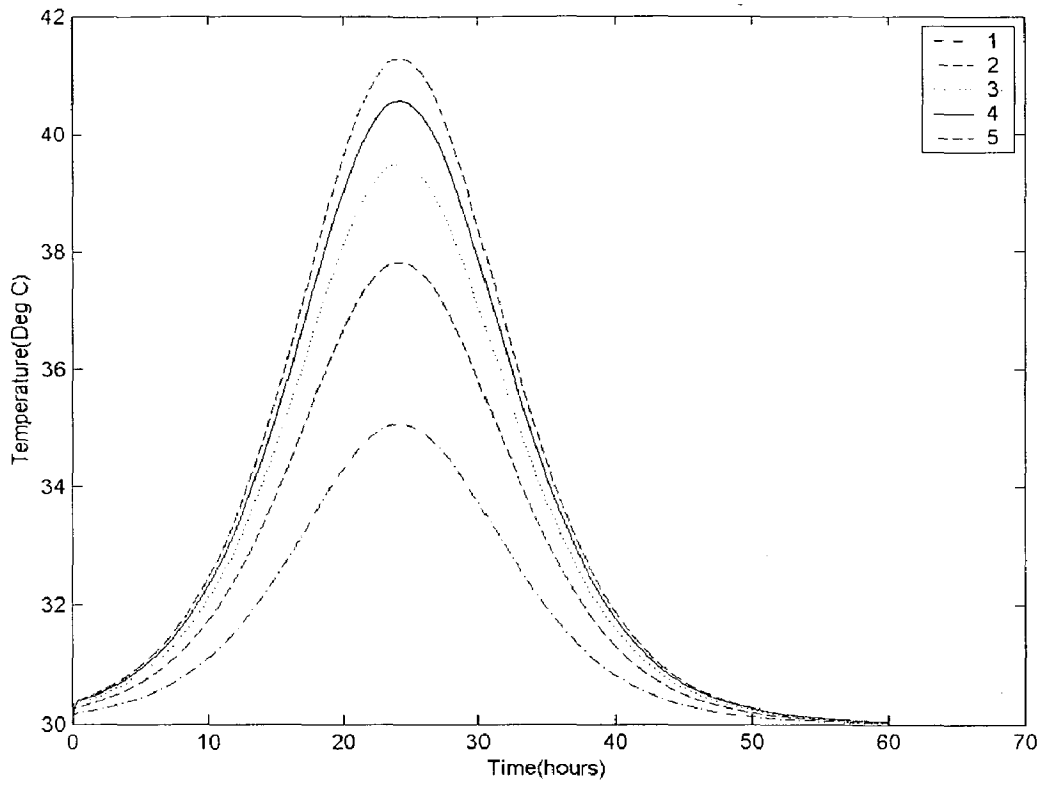


Figure 4.9 Plot of temperature vs. time for *Aspergillus niger*
 ($N=5, F_g=0, \mu=0.2, \alpha=0.2$)

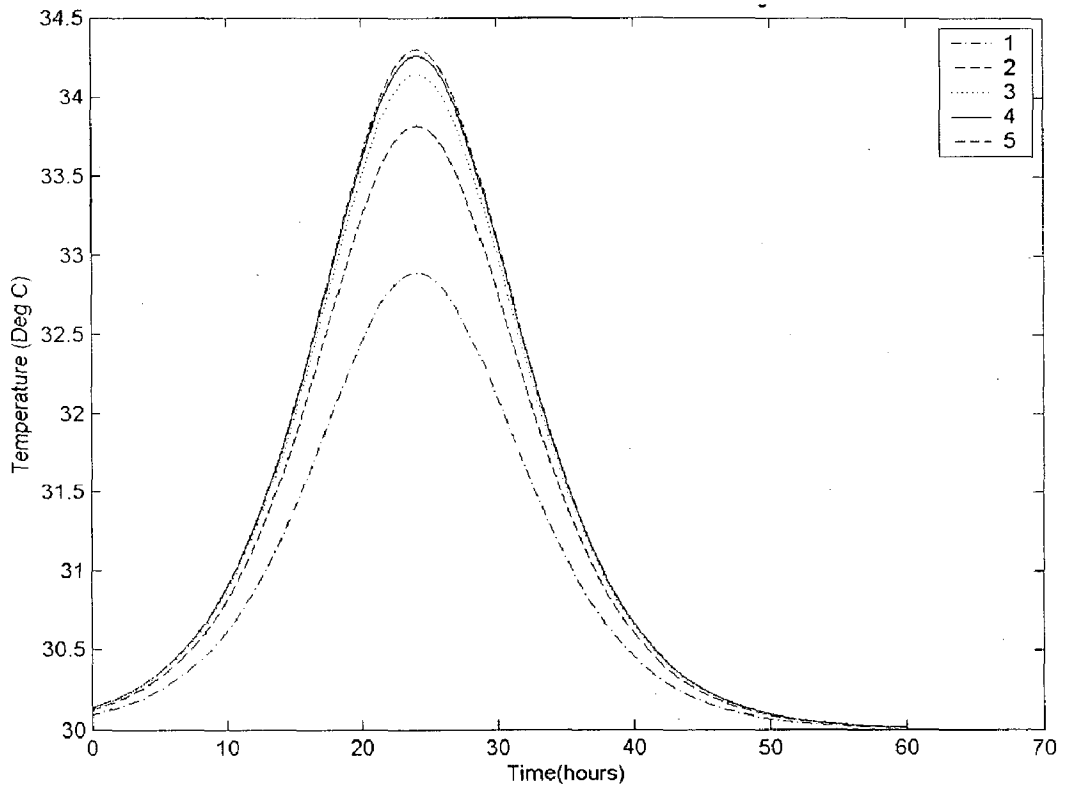


Figure 4.10 Plot of temperature vs. time for *Aspergillus niger*
 ($N=5, F_g=2, \mu=0.2, \alpha=0.2$)

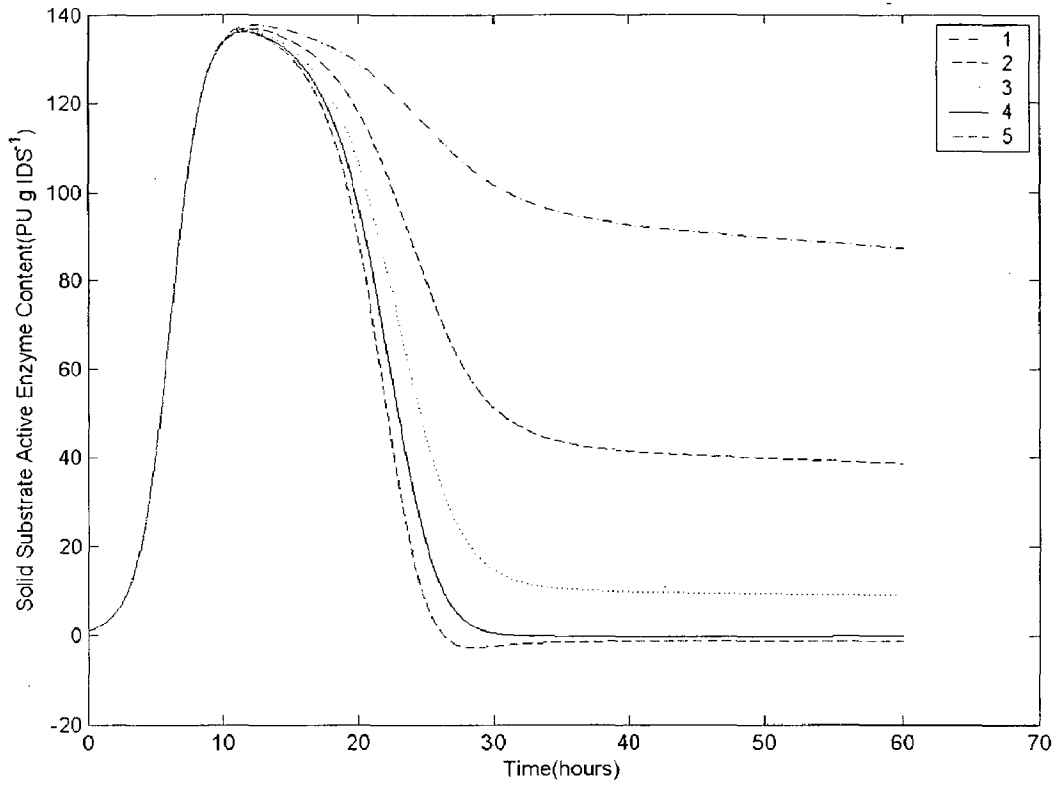


Figure 4.11 Plot of Solid Substrate Active Enzyme Content vs. time for *Aspergillus niger* ($N=5, F_g=0, \mu=0.2, \alpha=0.2$)

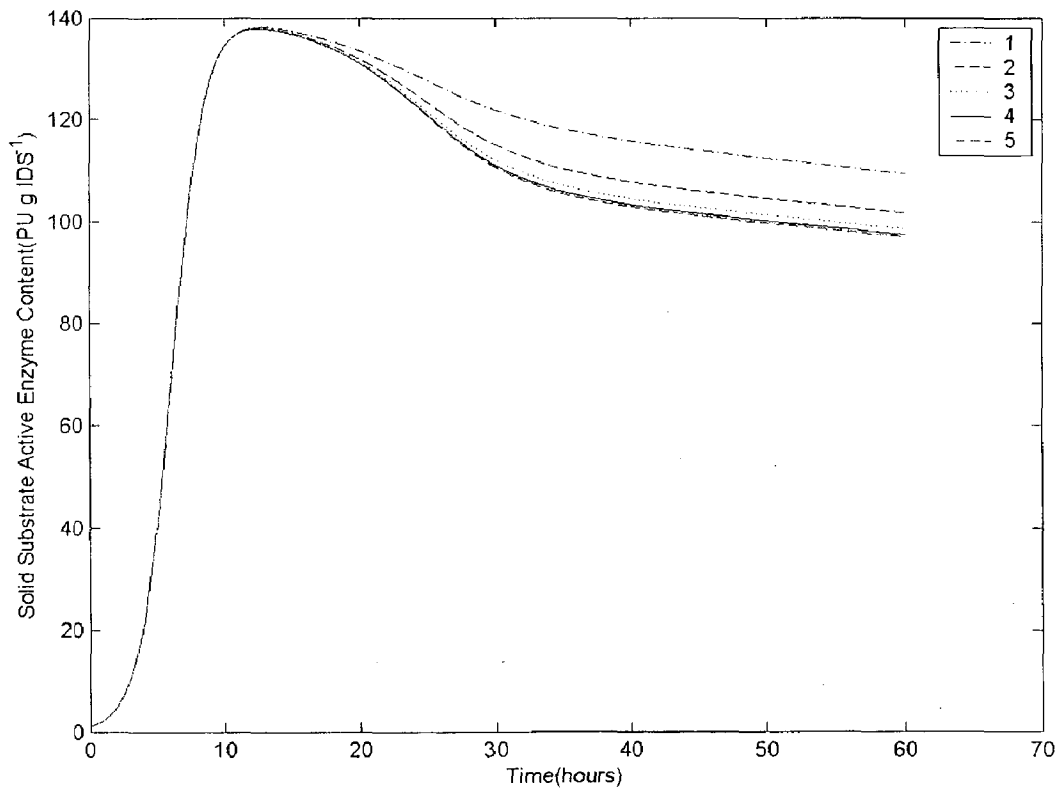


Figure 4.12 Plot of Solid Substrate Active Enzyme Content vs. time for *Aspergillus niger* ($N=5, F_g=2, \mu=0.2, \alpha=0.2$)

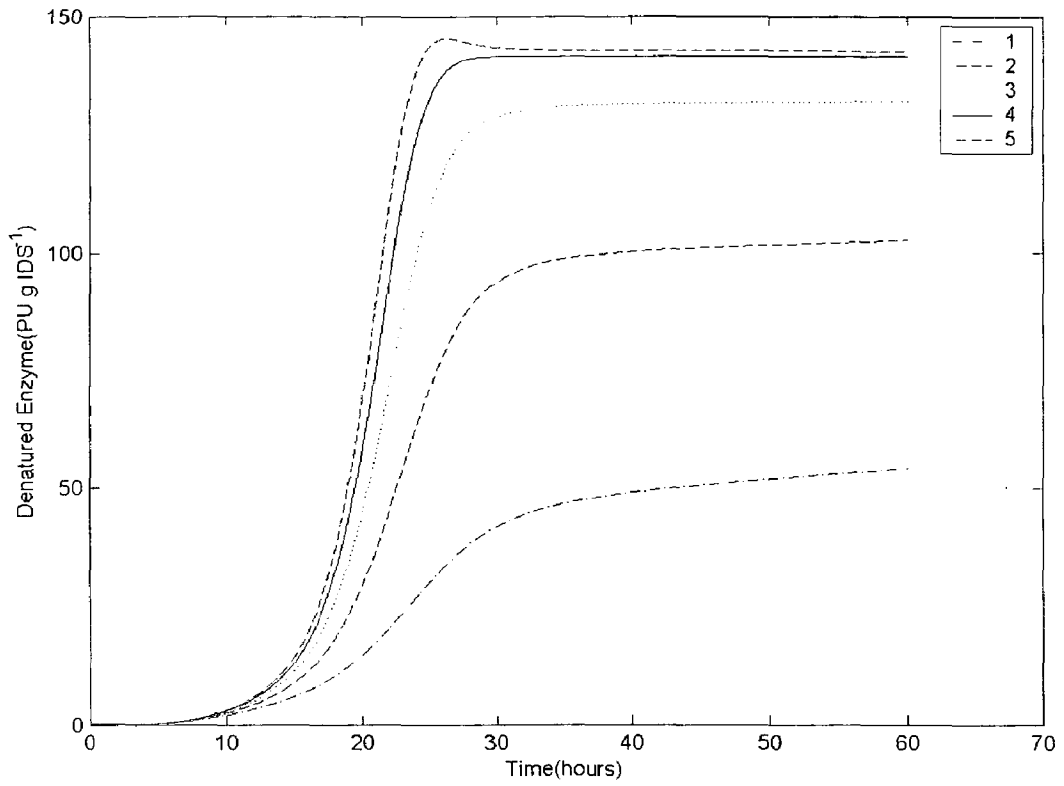


Figure 4.13 Plot of Denatured Enzyme vs. time for *Aspergillus niger*
 ($N=5, F_g=0, \mu=0.2, \alpha=0.2$)

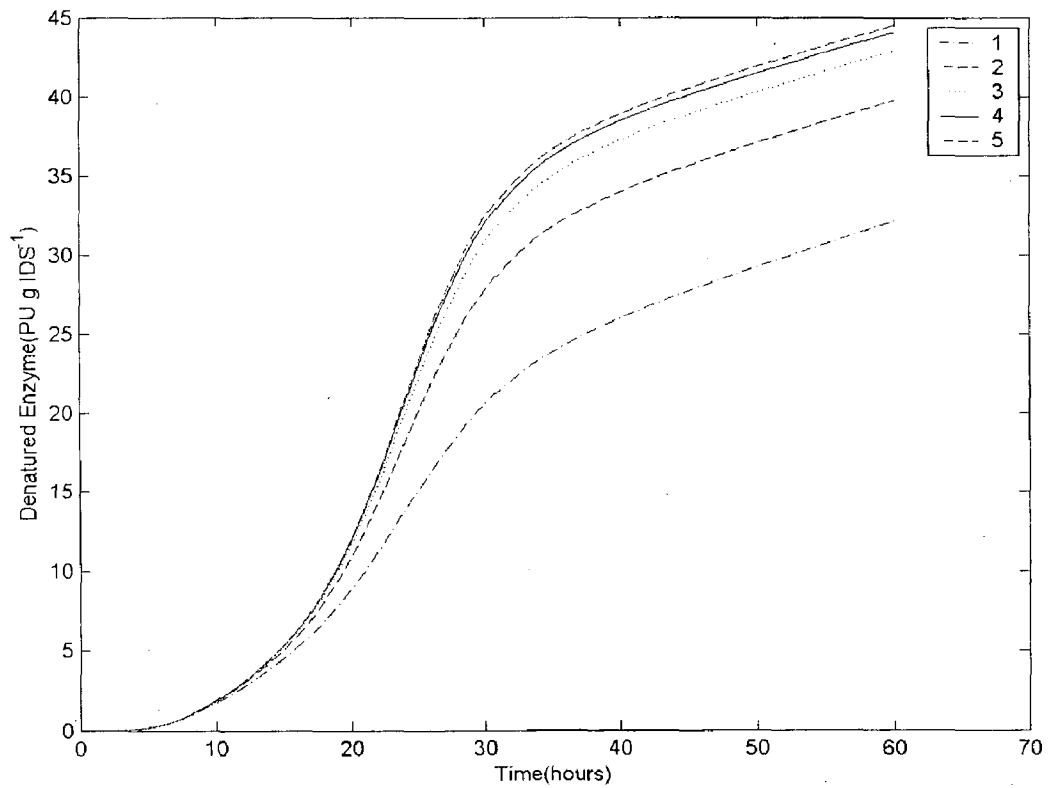


Figure 4.14 Plot of Denatured Enzyme vs. time for *Aspergillus niger*
 ($N=5, F_g=2, \mu=0.2, \alpha=0.2$)

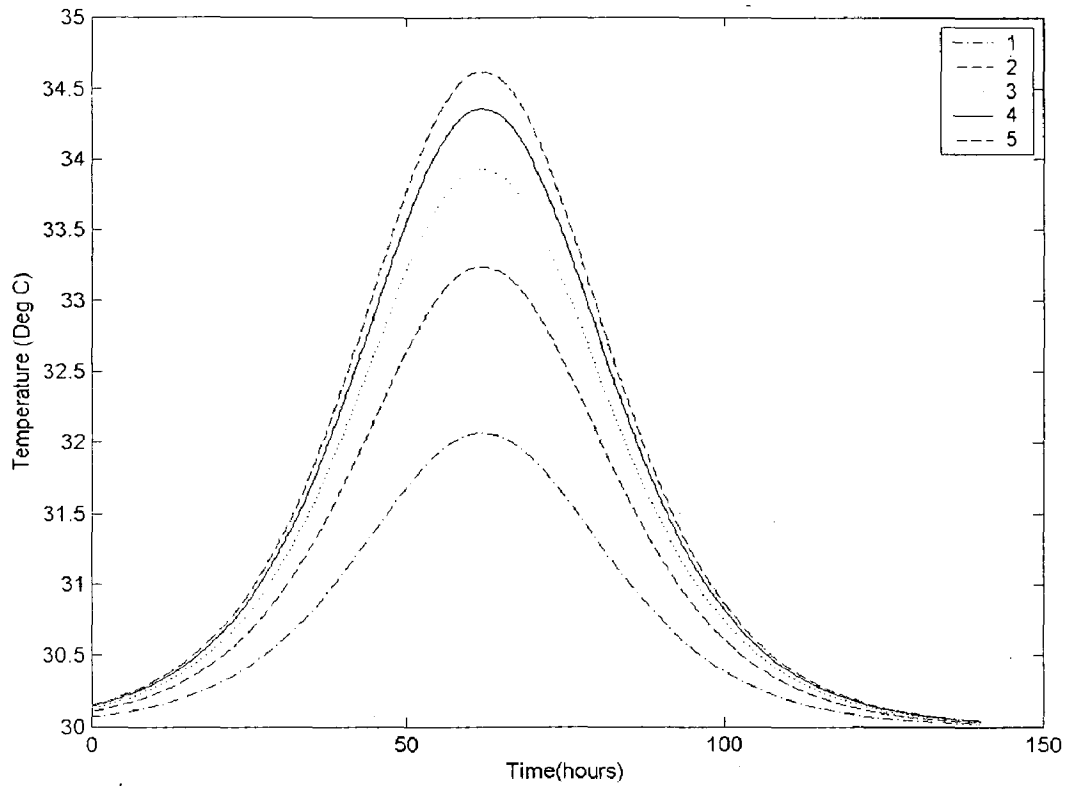


Figure 4.15 Plot of Temperature vs. time for *Penicillium fellutanum*
 ($N=5, F_g=0, \mu=0.0782, \alpha=0.0782$)

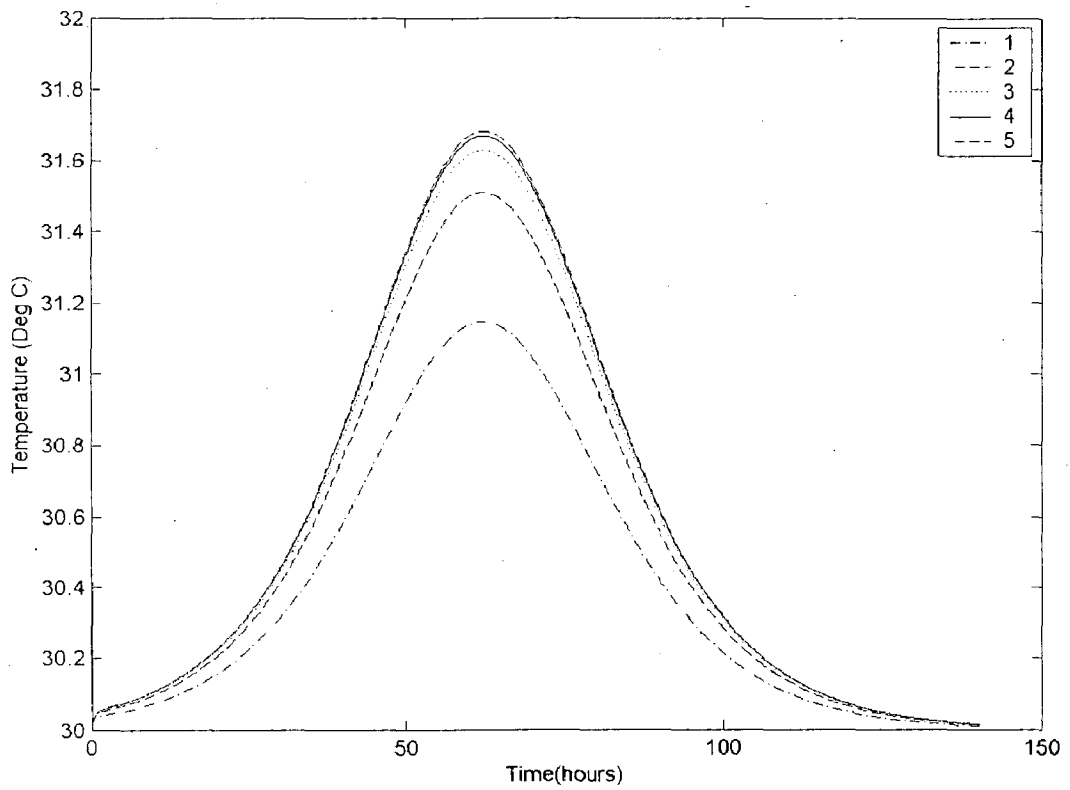


Figure 4.16 Plot of Temperature vs. time for *Penicillium fellutanum*
 ($N=5, F_g=2, \mu=0.0782, \alpha=0.0782$)

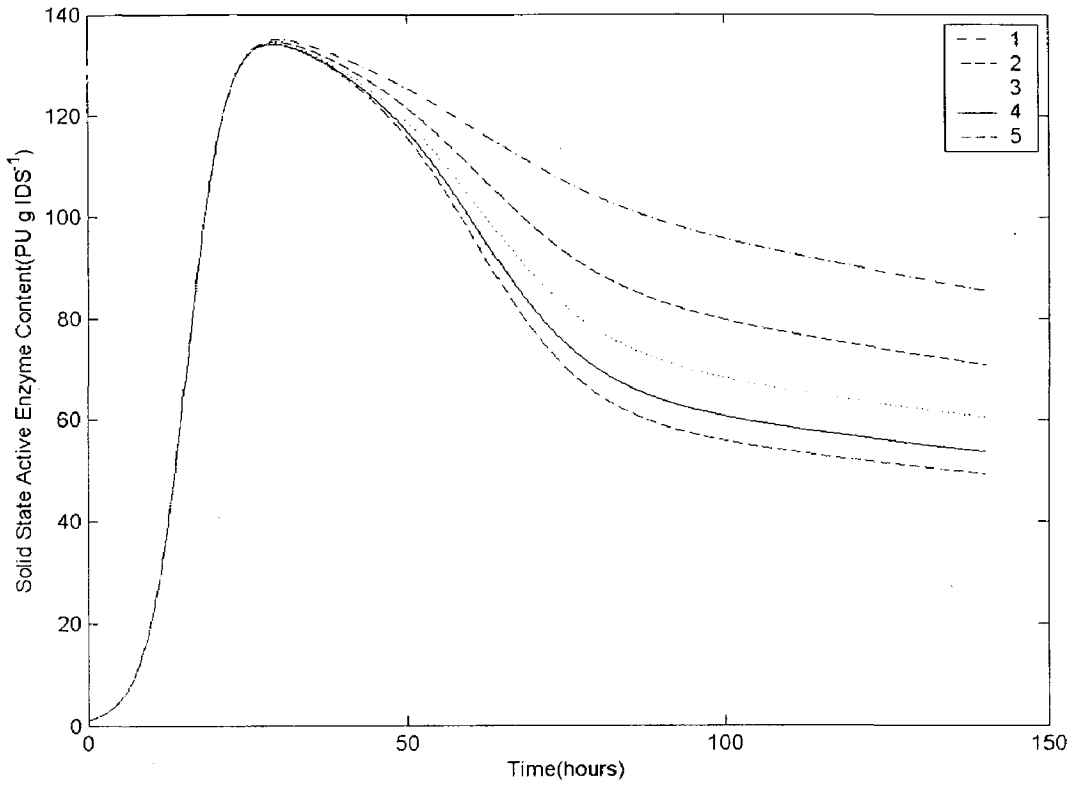


Figure 4.17 Plot of Solid State Active Enzyme Content vs. time for *Penicillium fellutanum* ($N=5, F_g=0, \mu=0.0782, \alpha=0.0782$)

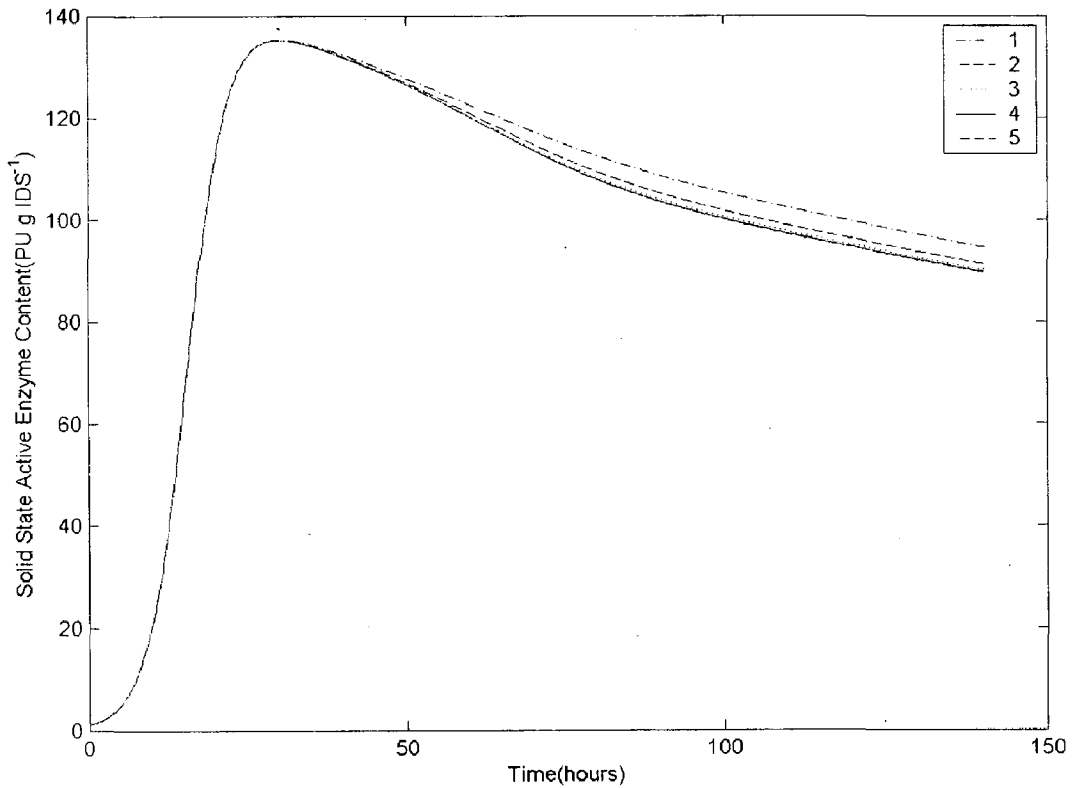


Figure 4.18 Plot of Solid State Active Enzyme Content vs. time for *Penicillium fellutanum* ($N=5, F_g=2, \mu=0.0782, \alpha=0.0782$)

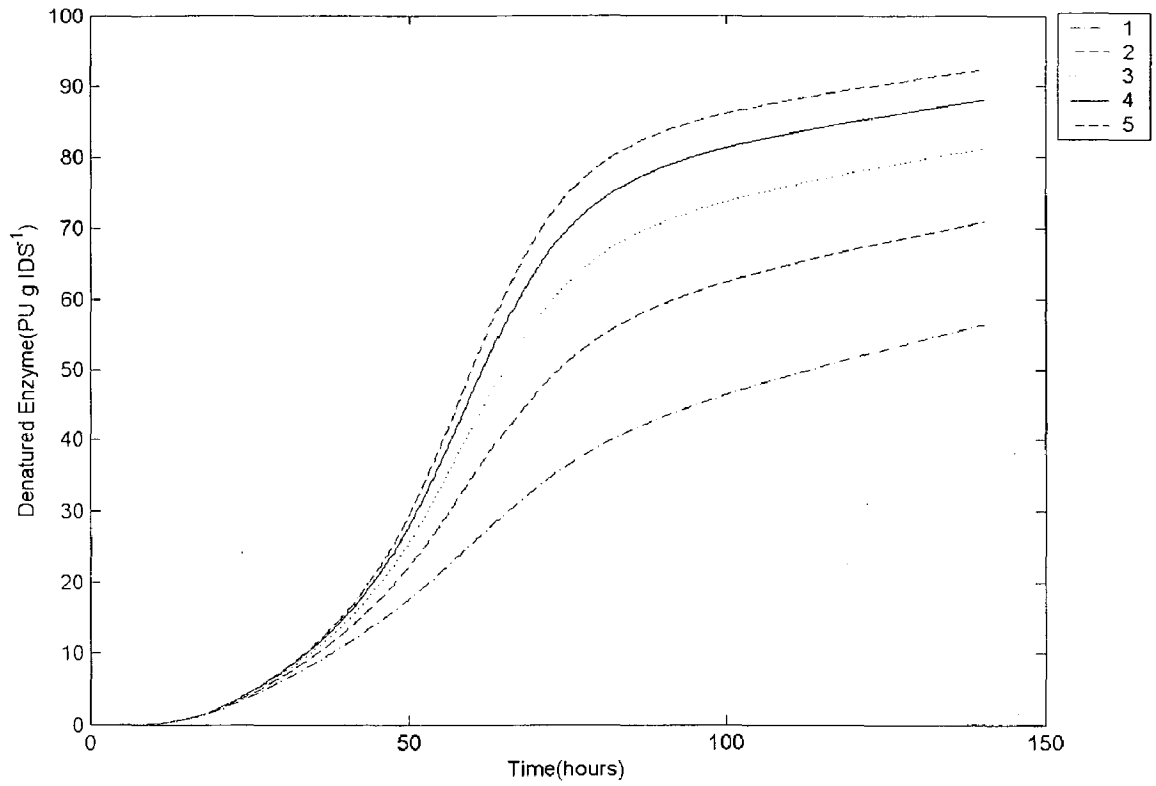


Figure 4.19 Plot of Denatured Enzyme vs. time for *Penicillium fellutanum*
 ($N=5, F_g=0, \mu=0.0782, \alpha=0.0782$)

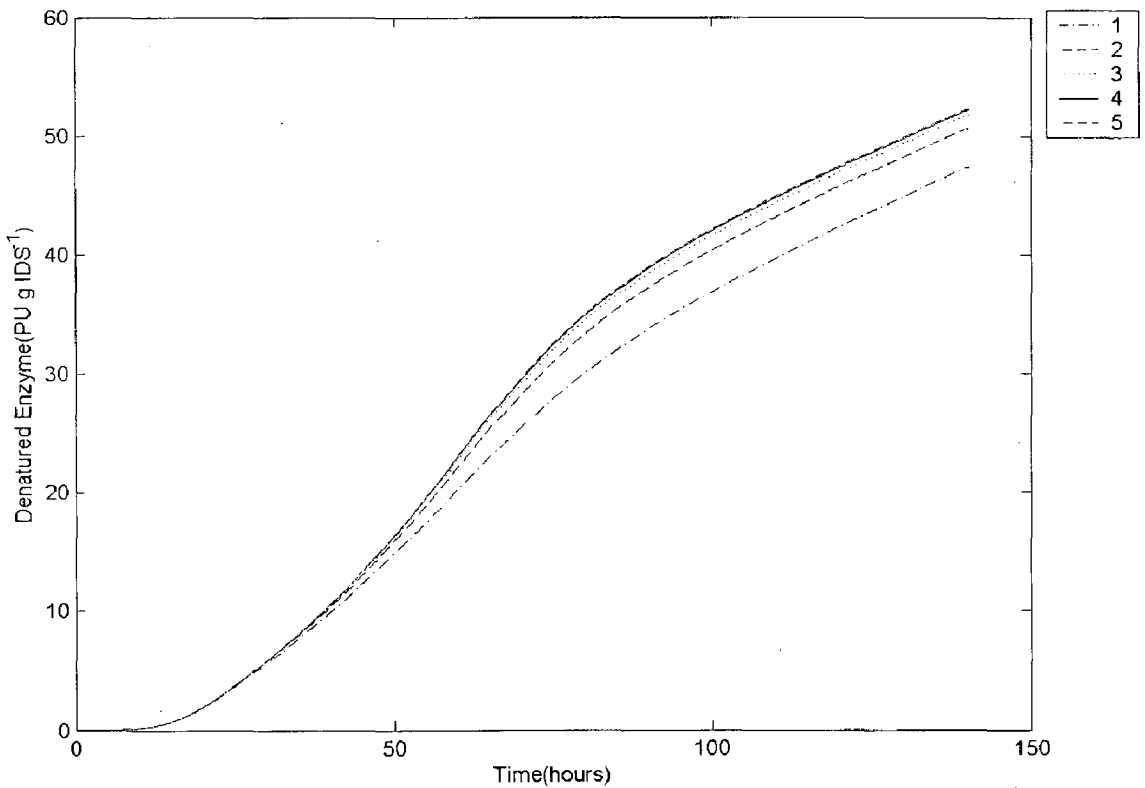


Figure 4.20 Plot of Denatured Enzyme vs. time for *Penicillium fellutanum*
 ($N=5, F_g=2, \mu=0.0782, \alpha=0.0782$)

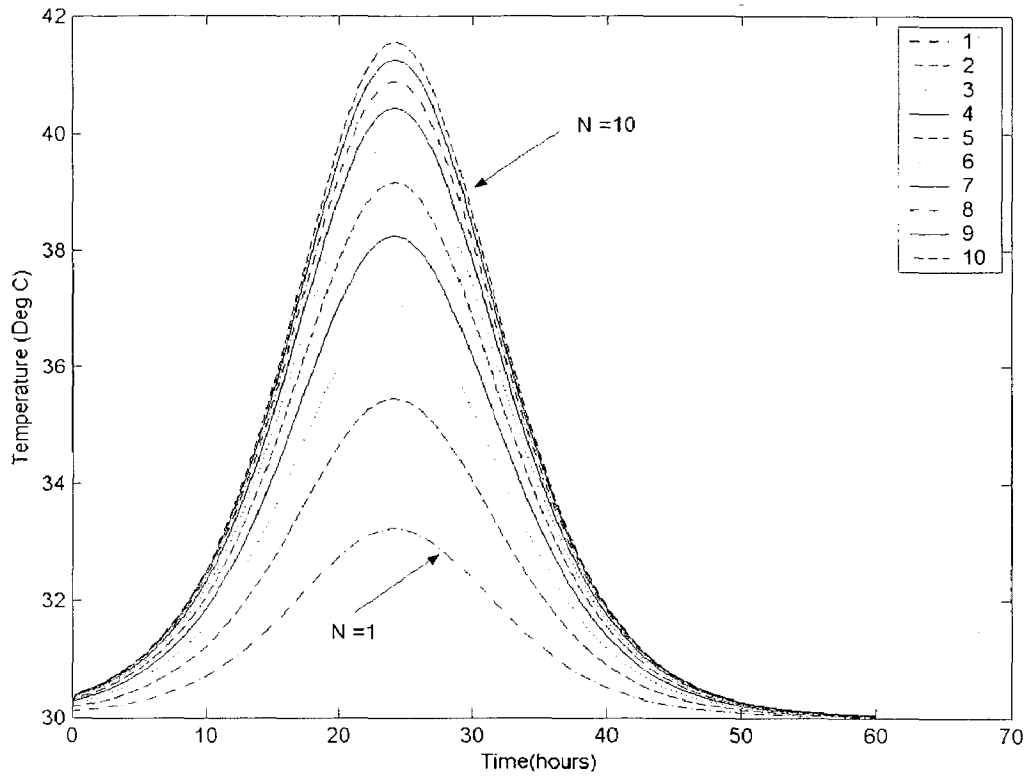


Figure 4.21 Plot of Temperature vs. time for Aspergillus niger
 ($N=10, F_g=0, \mu=0.2, \alpha=0.2$)

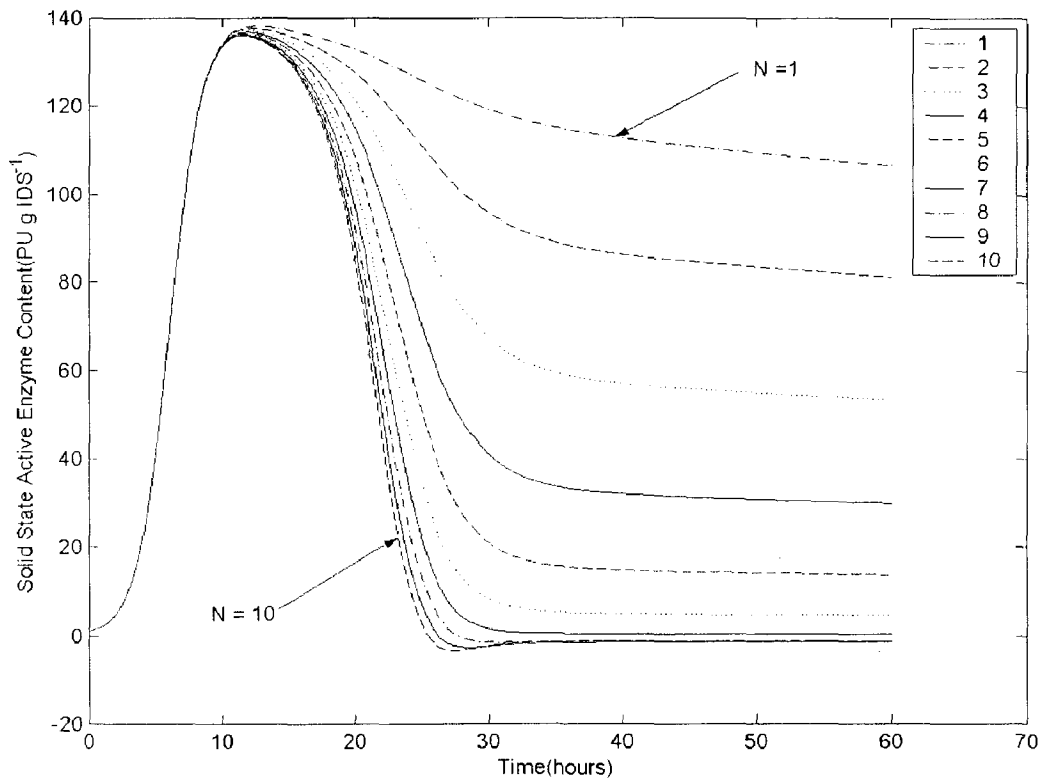


Figure 4.22 Plot of Solid State Active Enzyme Content vs. time for Aspergillus niger
 ($N=10, F_g=0, \mu=0.2, \alpha=0.2$)

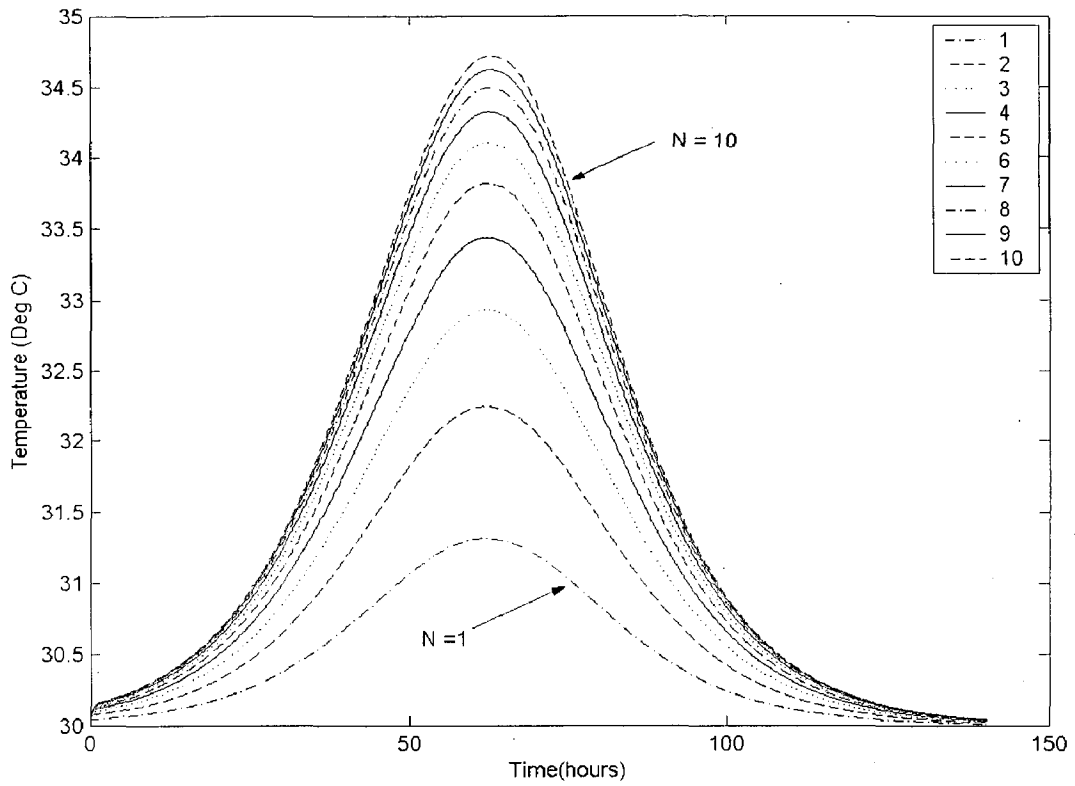


Figure 4.25 Plot of Temperature vs. time for *Penicillium fellutanum*
 ($N=10, F_g=0, \mu=0.0782, \alpha=0.0782$)

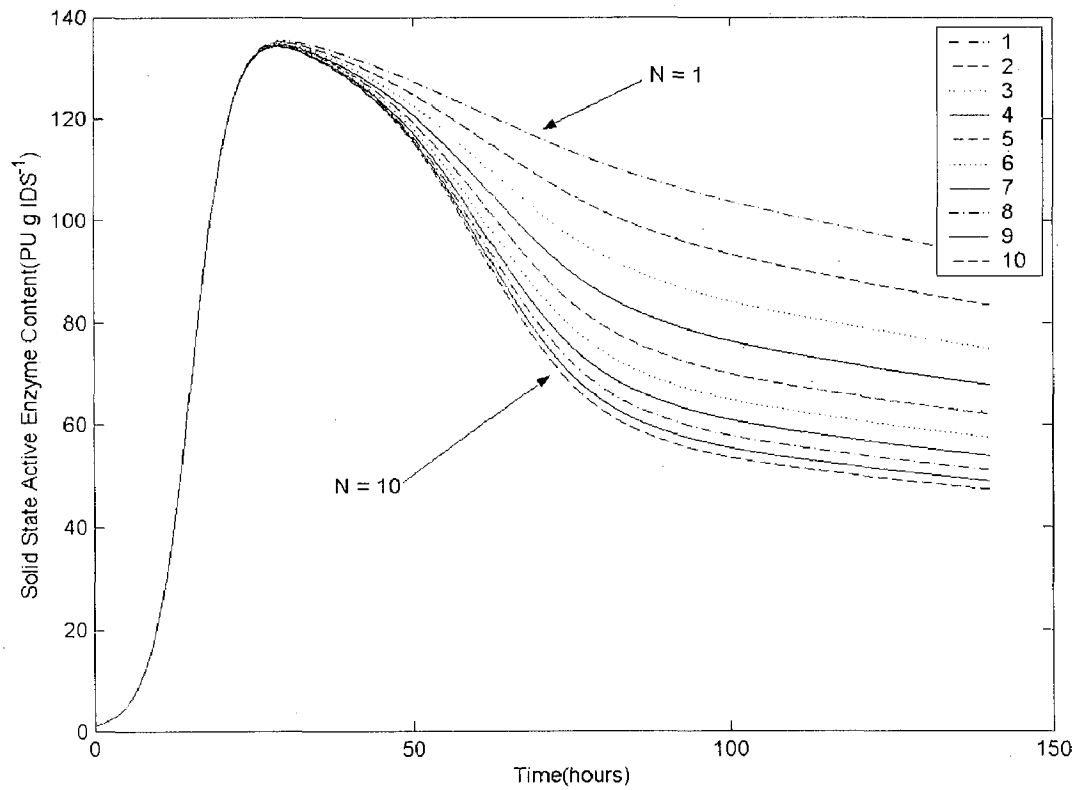


Figure 4.26 Plot of Solid State Active Enzyme Content vs. time for *Penicillium fellutanum* ($N=10, F_g=0, \mu=0.0782, \alpha=0.0782$)

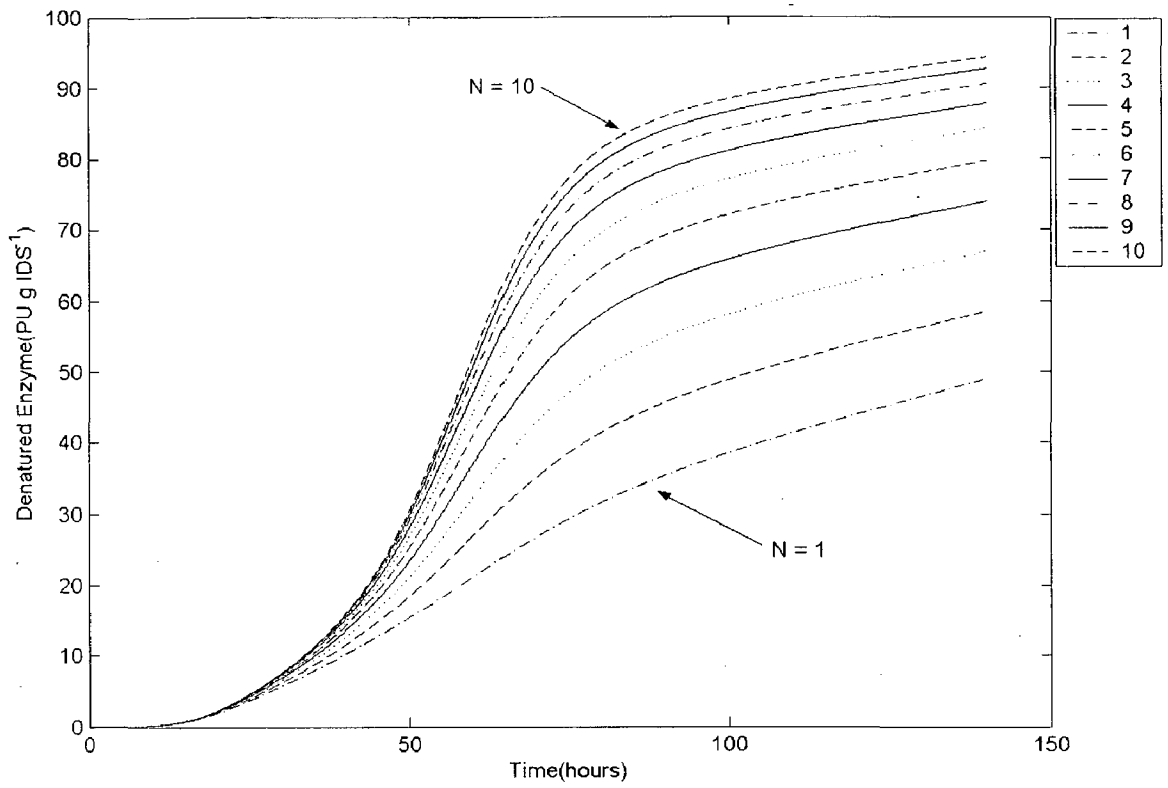


Figure 4.27 Plot of Denatured Enzyme vs. time for Penicillium fellutanum
 ($N=10, F_g=0, \mu=0.0782, \alpha=0.0782$)

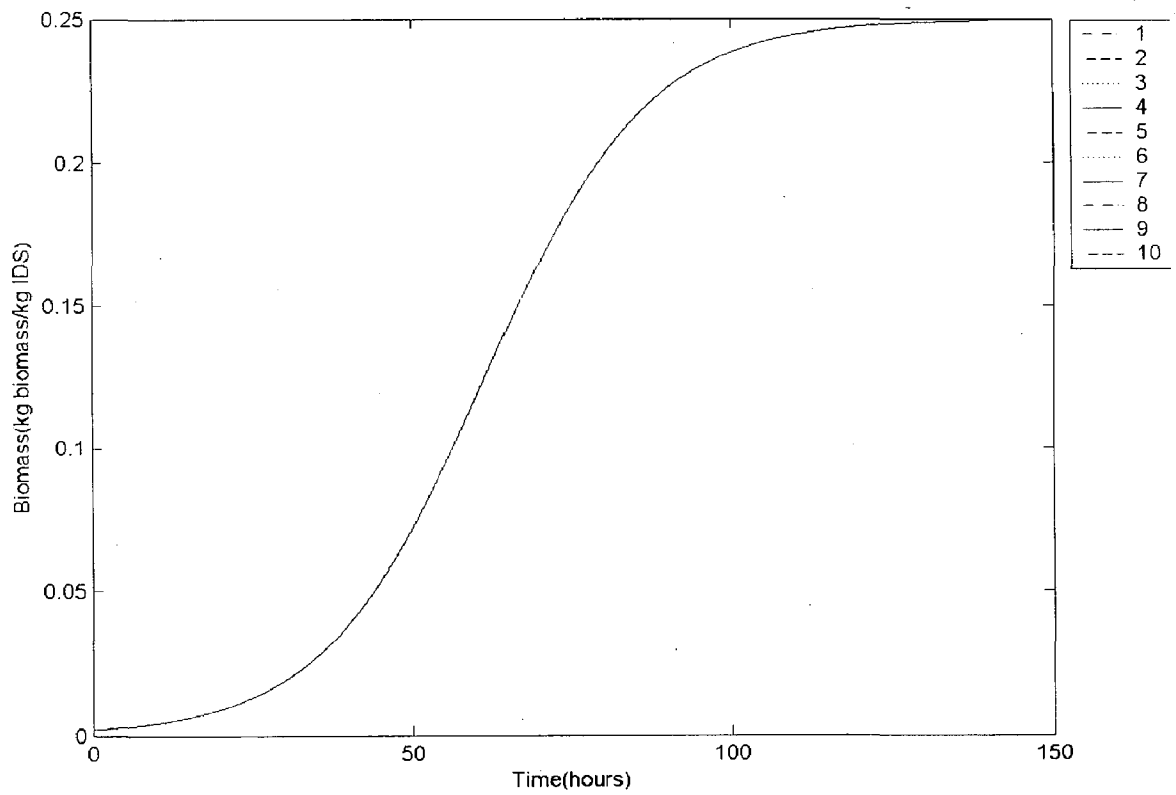


Figure 4.28 Plot of Biomass Content vs. time for Penicillium fellutanum
 ($N=10, F_g=0, \mu=0.0782, \alpha=0.0782$)

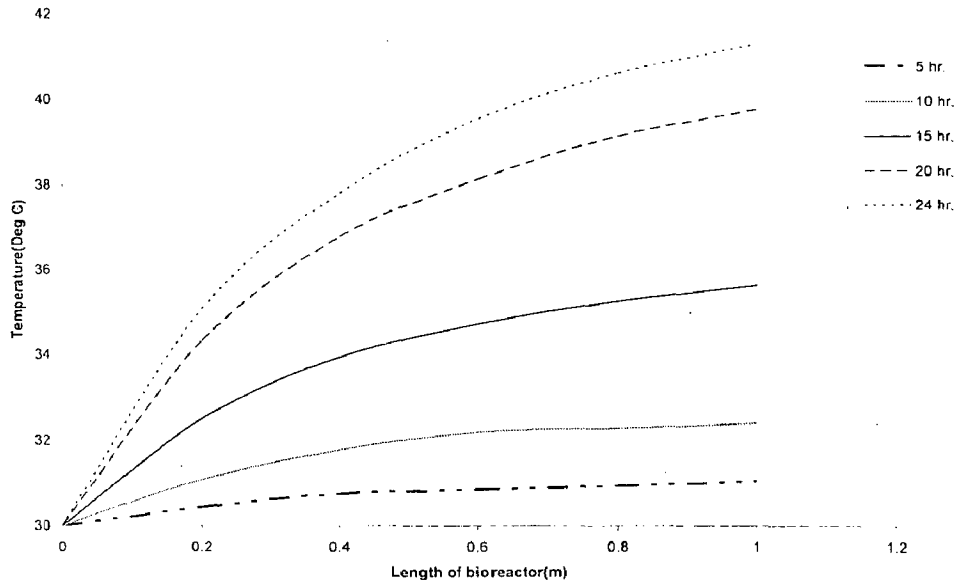


Figure 4.29 Plot of Temperature vs. Length of Bioreactor for *Aspergillus niger* ($F_g=0, \mu=0.2, \alpha=0.2$)

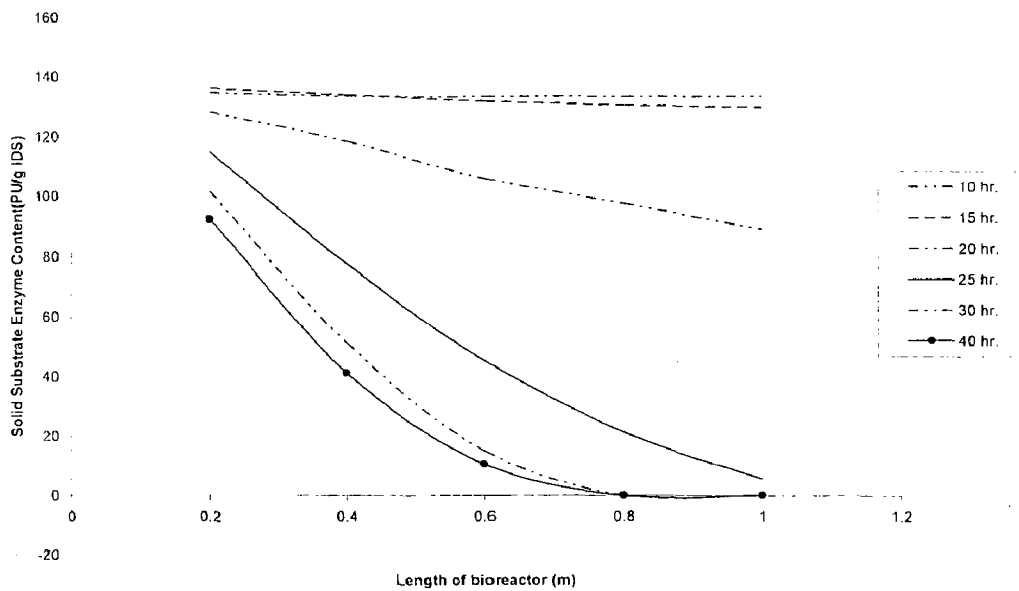


Figure 4.30 Plot of Solid State Active Enzyme Content vs. length of bioreactor for *Aspergillus niger* ($F_g=0, \mu=0.2, \alpha=0.2$)

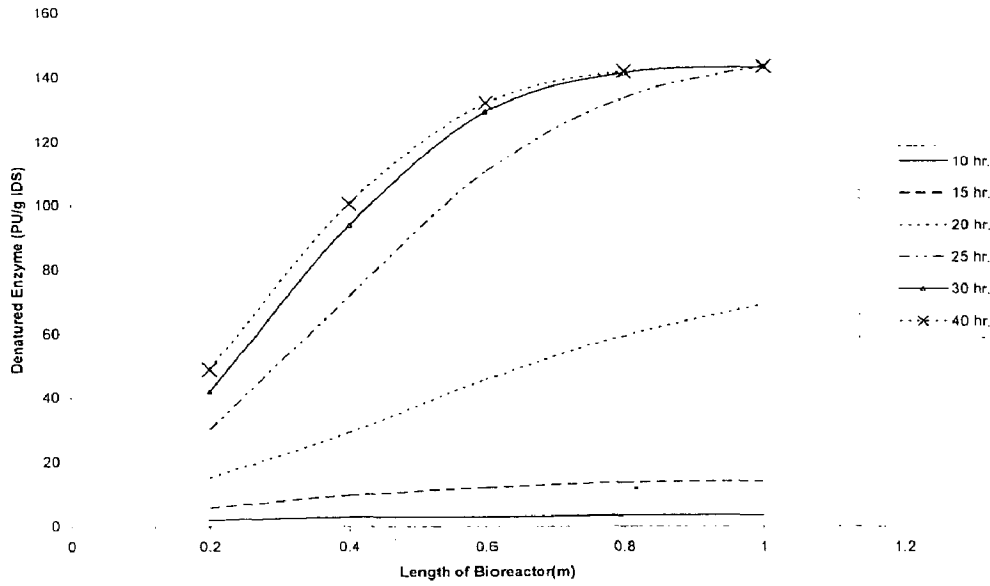


Figure 4.31 Plot of Denatured Enzyme vs.length of bioreactor for *Aspergillus niger*. ($F_g=0, \mu=0.2, \alpha=0.2$)

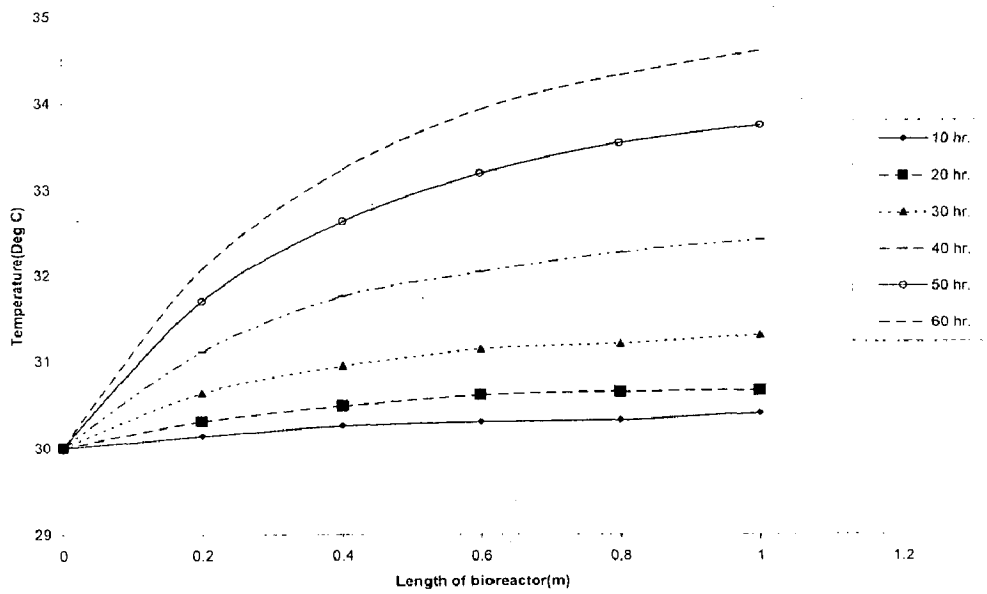


Figure 4.32 Plot of Temperature vs.length of bioreactor for *Penicillium fellutanum* ($F_g=0, \mu=0.0782, \alpha=0.0782$)

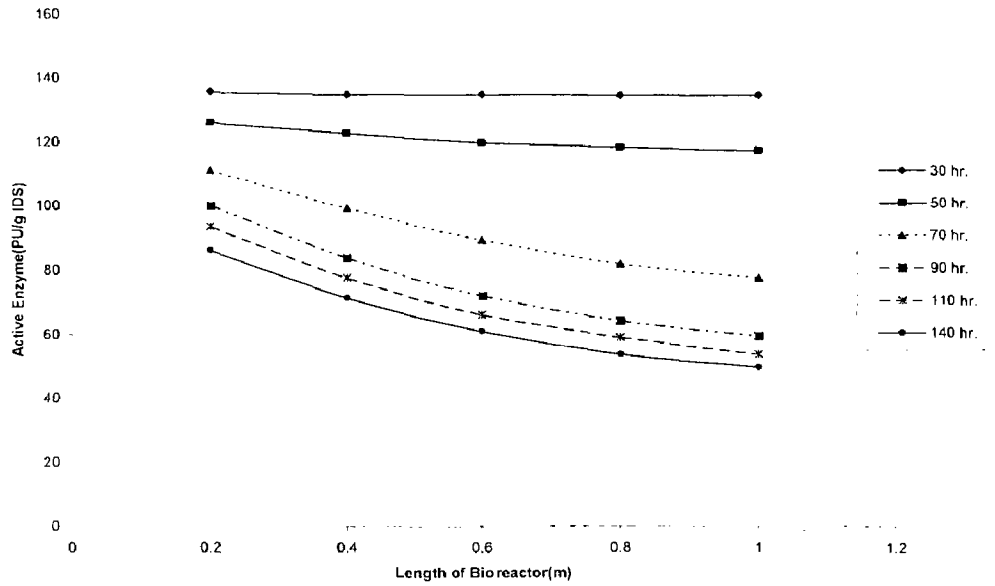


Figure 4.33 Plot of Solid Substrate Active Enzyme Content vs. Length of Bioreactor for *Penicillium fellutanum* ($F_g=0, \mu=0.0782, \alpha=0.0782$)

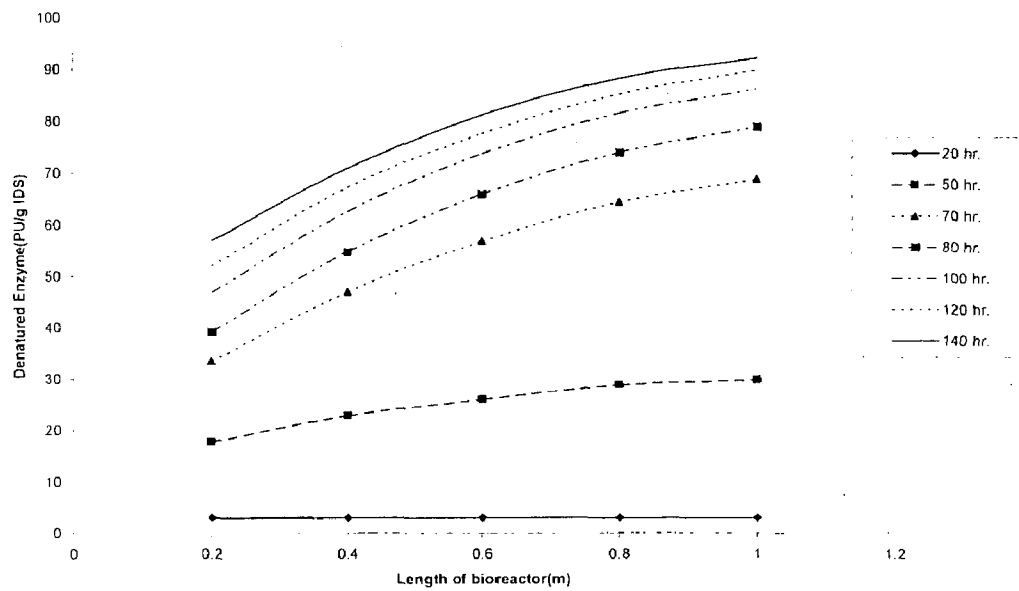


Figure 4.34 Plot of Denatured Enzyme vs. length of bioreactor for *Penicillium fellutanum* ($F_g=0, \mu=0.0782, \alpha=0.0782$)

4.1.5 Validation of the Mathematical Model

The mathematical model was validated against the experimental data of Ghildyal et al. (1994). They had conducted experiments in a 150 mm. diameter, 345 mm. high packed bed bioreactor wherein the production of amyloglucosidase enzyme was done by *Aspergillus niger* on Wheat Bran.

In their study, Ghildyal et al. did not use a water jacket to remove heat generated during the solid state fermentation process, Hence in the pseudohomogeneous model developed the term which involves the transfer of heat to the water jacket is neglected for the case of validation. Thus equation 3.20 is employed as given below

$$\frac{dT_n}{dt} = \frac{\left[F_a C_{pa} (T_{n-1} - T_n) + F_a \lambda (h_a |_{T_{n-1}} - h_a |_{T_n}) + F_a C_{pv} (h_a |_{T_{n-1}} T_{n-1} - h_a |_{T_n} T_n) + \left(\frac{B}{n} \right) Y_Q \frac{dX}{dt} - h \left(\frac{A}{n} \right) (T_n - T_w) \right]}{\left(\frac{B}{n} \right) (1+W) C_{pb}} \quad (3.20)$$

$$\frac{dT_n}{dt} = \frac{\left[F_a C_{pa} (T_{n-1} - T_n) + F_a \lambda (h_a |_{T_{n-1}} - h_a |_{T_n}) + F_a C_{pv} (h_a |_{T_{n-1}} T_{n-1} - h_a |_{T_n} T_n) + \left(\frac{B}{n} \right) Y_Q \frac{dX}{dt} \right]}{\left(\frac{B}{n} \right) (1+W) C_{pb}} \quad (4.1)$$

Sangsurasak and Mitchell (1998) had developed a two dimensional mathematical model (2.2.3) and had validated their results using the data of Ghildyal et al.. The model was solved by Orthogonal Collocation, wherein Jacobi Polynomials were used to discretize the spatial coordinates as a two dimensional axisymmetric problem.

Ghildyal et al. measured the temperature –time profiles at the centre of the bed (i.e. at 170 mm.) for different air flow rates. The temperature –time

profiles obtained for different air flow rates as solved by the N-Tanks in Series approach are shown in figs. 4.35,4.37,4.39 for 5,15 and 25 lpm flow rate of air respectively.

As a comparison the results obtained by the solution of two dimensional model by Sangsurasak and Mitchell are shown in the figs. 4.36,4.38 and 4.40 for 5,15 and 25 lpm respectively.

From the figures it can be clearly observed that there is a good agreement between the results of the N-Tanks in Series approach and those of Ghildyal et al. for a wide range of superficial velocities. The results obtained are in excellent concordance with those obtained by Sangsurasak and Mitchell(1998) ,who had attributed the differences arising in the results due to two primary factors in their paper:-

- i) Ghildyal et al.(1994) did not determine many parameters which were necessary for the solution of the two dimensional model.
- ii) Many parameters used for the solution of their two-dimensional model are those of Saucedo-Castañeda et al. (1990).Hence the difference in substrates might affect growth kinetics and heat transfer properties.

The solution of the model equation by N-Tanks in Series approach assumes significance as it can be inferred that it is not always necessary to go for computational techniques like Orthogonal Collocation which are difficult to implement.

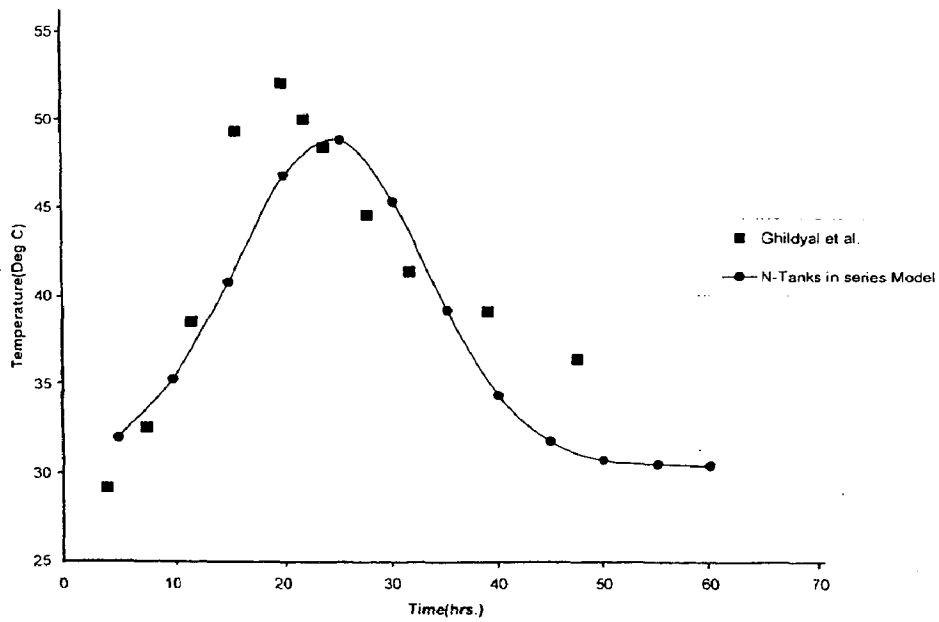


Figure 4.35 Validation of the N-Tanks in Series Model with the data of Ghildyal et al.(1994) for a flow rate of 5 lpm for *Aspergillus niger*

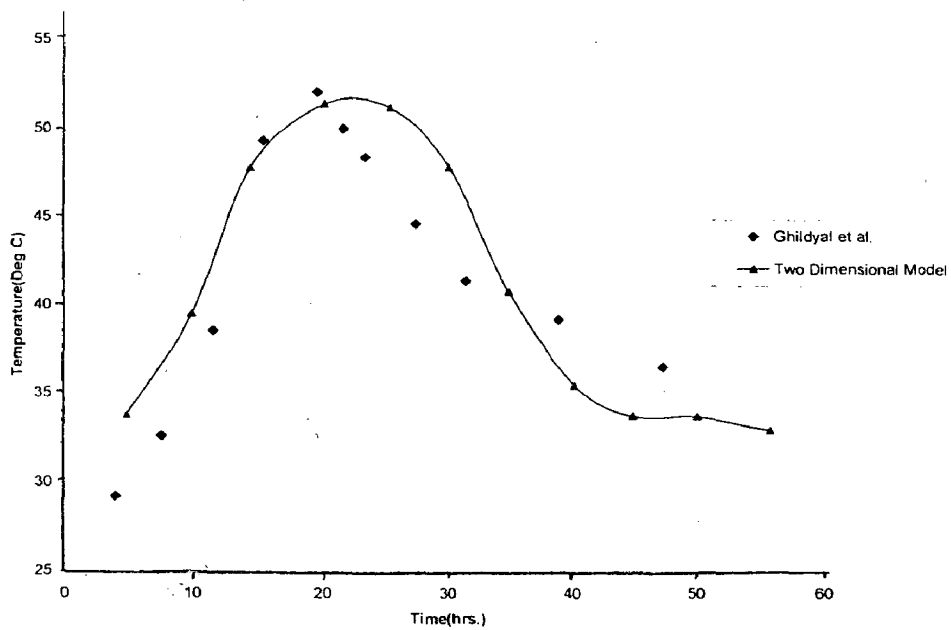


Figure 4.36 Validation of the Two Dimensional Model (Sangsurasak and Mitchell, 1998) with the data of Ghildyal et al.(1994) for a flow rate of 5 lpm for *Aspergillus niger*

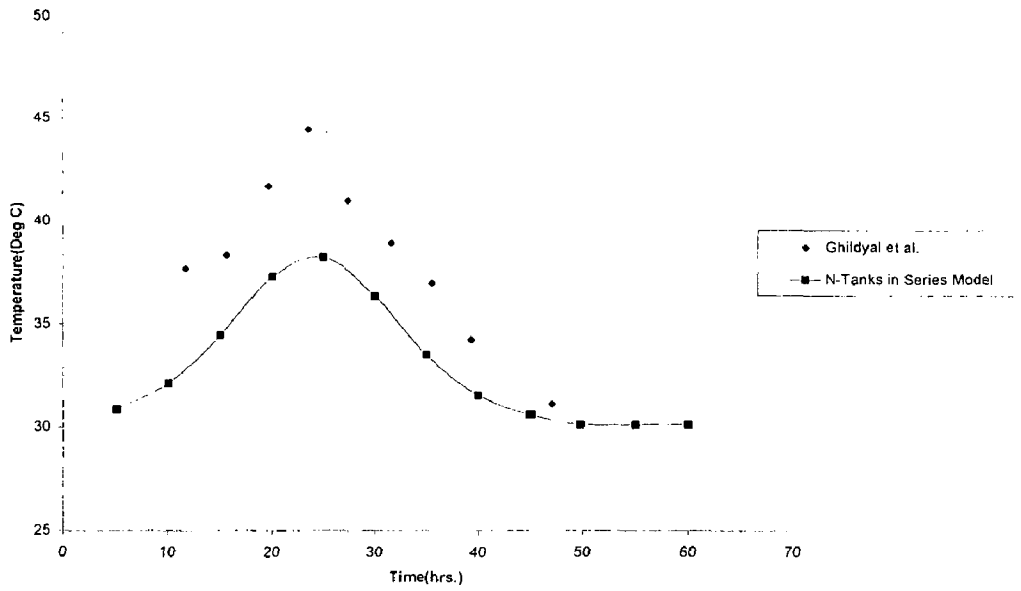


Figure 4.37 Validation of the N-Tanks in Series Model with the data of Ghildyal et al.(1994) for a flow rate of 15 lpm for *Aspergillus niger*

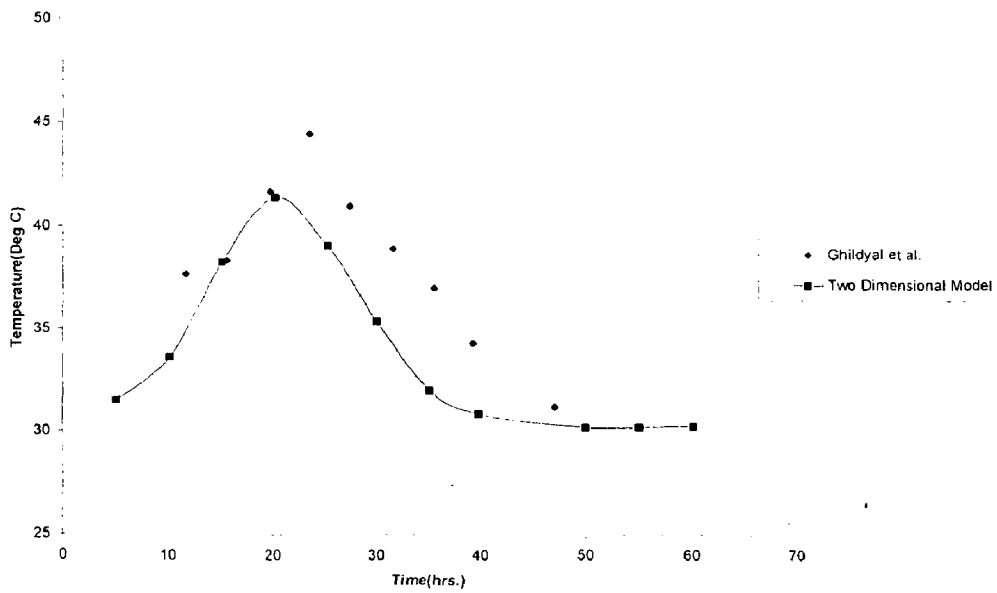


Figure 4.38 Validation of the Two Dimensional Model (Sangsurasak and Mitchell, 1998) with the data of Ghildyal et al.(1994) for a flow rate of 15 lpm for *Aspergillus niger*

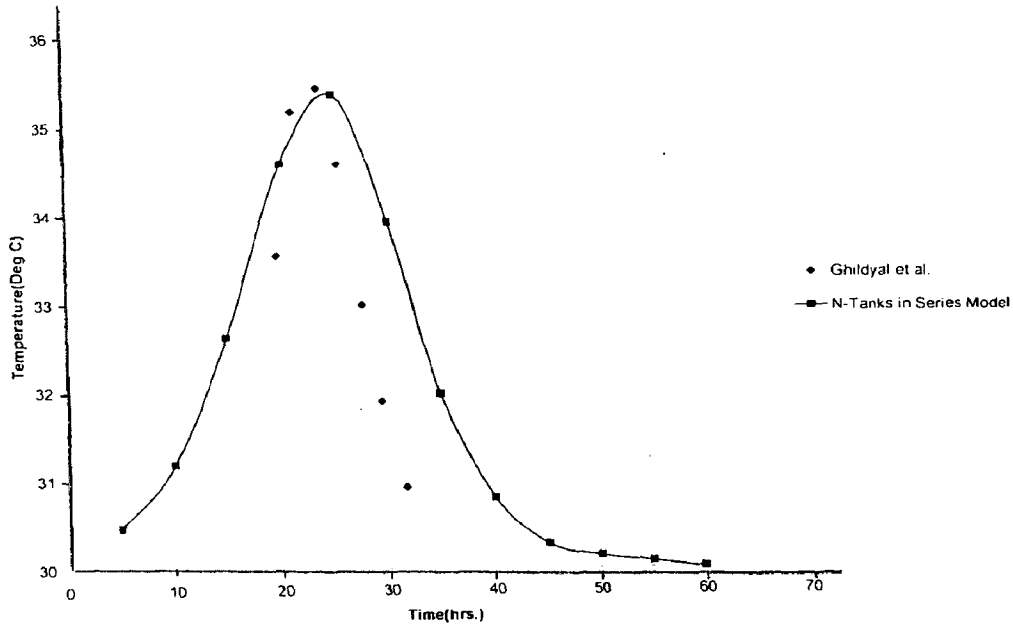


Figure 4.39 Validation of the N-Tanks in Series Model with the data of Ghildyal et al.(1994) for a flow rate of 25 lpm for *Aspergillus niger*

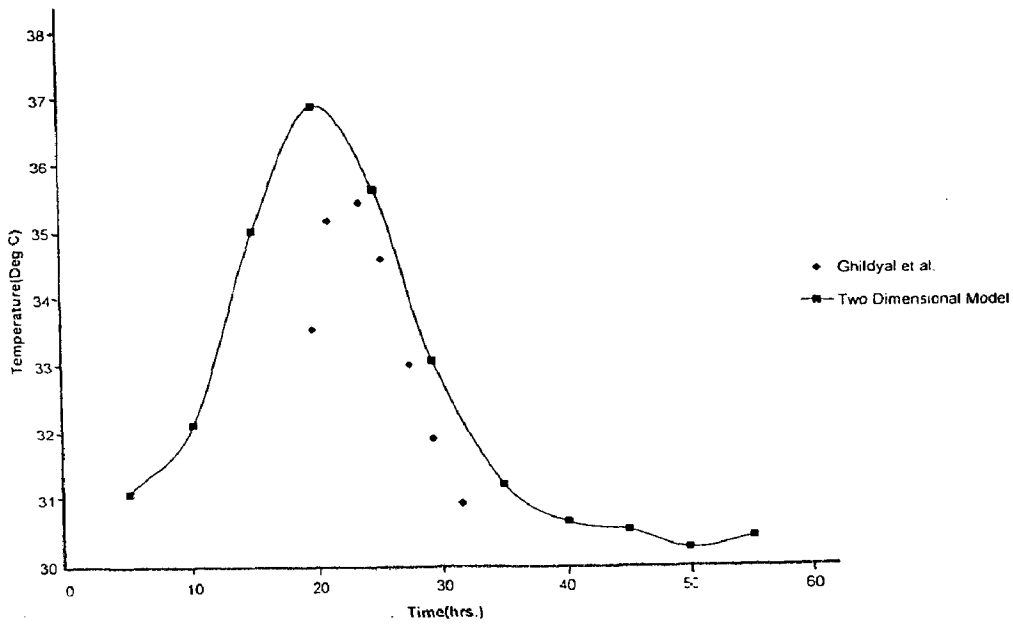


Figure 4.40 Validation of the Two Dimensional model (Sangsurasak and Mitchell, 1998) with the data of Ghildyal et al.(1994) for an air flow rate of 25 lpm for *Aspergillus niger*

4.2 RESULTS FROM THE TWO-PHASE MODEL

The two phase dynamic model was developed by Von Meien and Mitchell (2002). In it they had considered the substrate bed (solid phase) and air (gaseous phase) as two distinct phases, and had applied mass and energy balances on them. In this section the main aim is to extend the application of N-Tanks in Series approach to Packed Bed Solid State Fermentation Bioreactors modelled as two phase systems.

4.2.1 Computational Experiences

The system of model equations developed using the N-Tanks in Series approach (3.76-3.81) constituted a set of ordinary differential equations which was highly stiff in nature. This experience of ours is confirmed by the fact that Von Meien and Mitchell (2002) had used DASSL routine to solve the set of equations in their model, which is a Ordinary Differential Equation solver used generally for stiff ordinary differential equations.

4.2.2 Profiles of temperature and water content of solid and gaseous phases, biomass and solid phase concentration with time

The profiles of temperature and water content of solid phase; temperature and water content of gaseous phase, solid phase and biomass concentration have been obtained by varying the number of tanks ,N:- N=1(Fig. 4.41-4.46); N=3(Figs. 4.47-4.52) and for N=5(Fig. 4.53-4.58).The inferences which could be drawn from the obtained results:-

1. The temperature of solid phase for N=1(Fig. 4.41) at 6 hrs. is 49.2 °C .The temperature of solid phase profiles for N=3(Fig. 4.47) and N=5(Fig. 4.53) indicate that about 40% of the solid bed experiences a temperature less than 45°C.
2. The water content of the solid phase vs. time profile emphasizes the importance of phenomena of drying. Significant drying occurs between 5 hours and 7.3 hrs. where the water content of the whole bed reduces from an initial value of 0.3325 kg water/kg dry solid to 0.157 kg water/kg dry solid in 80%of the solid bed. The lower 20% height of the bed in contact with moisturized air attains a value of 0.169 kg water/kg dry solids (Fig. 4.54).

3. The maximum reduction of concentration of the dry solids was observed in the lower 20% of the packed bed. At the end of 12 hours, the solid phase concentration reduced to 152 kg/m^3 from an initial solid phase concentration of 195 kg/m^3 (Fig. 4.49 and 4.55). If only $N=1$ (Fig. 4.43) is considered to represent the situation in the bioreactor then according to it a solid phase concentration of 162 kg/m^3 should prevail in the whole bioreactor, whereas Fig. 4.49 and Fig. 4.53 show that about 80% of the bed has a solid concentration above 162 kg/m^3 .
4. The maximum growth of biomass occurred in the lower 20% of the bed (Fig. 4.50 and 4.56). This maximal growth of biomass could be the possible reason for the maximum decrease in the dry solid concentration in the lower 20% of the packed bed.
5. The temperature of the gaseous phase is varying with the height of the bioreactor as indicated by the time profiles for $N=3$ and $N=5$ (Fig. 4.51 and 4.57 respectively), instead of the existence of a single temperature as indicated by $N=1$ (Fig. 4.45).
6. The water content of the gaseous phase also indicates variation with height. The peak water content of the gaseous phase, which is $0.075 \text{ kg water/kg dry air}$ (Fig. 4.46), occurs at 5.2 hrs., if $N=1$ is considered as the situation representing the whole bioreactor. The plot for $N=5$, (Fig. 4.58) indicates the occurrence of the peak water content at about 4.2 hrs. Through 80% the packed bed; the air passing through the bioreactor has a water content lower or equal to $0.075 \text{ kg water/kg dry air}$. Therefore all the air passing through the bioreactor does not have the same water content marked variations occur from $0.049 \text{ kg water/kg dry air}$ to $0.082 \text{ kg water/kg dry air}$ are present (Fig. 4.58).

A major conclusion that can be drawn from the above discussion is that for understanding the variation of variables with height the N-Tanks in Series Model can give important insights. The results of the two phase model are especially important since the water content in the solids may drop down to levels which may retard growth.

4.2.3 Comparison of the results from N-Tanks in Series approach with Von Meien and Mitchell (2002)

Von Meien and Mitchell (2002) had reported that the solid phase temperature at the outlet was near maximal at 12 hours. The application of N-Tanks in series model, using the same model parameters as those by Von Meien and Mitchell (2002), the solid phase temperature at the outlet peaked at 4.5 hours. Fig. 4.59 shows the temperature vs. fractional axial distance or fractional height obtained by N-Tanks in Series approach and Fig 4.60 shows the profiles obtained by Von Meien and Mitchell (2002). It can be clearly noticed that there is an excellent agreement between the peak temperatures of solid except the time at which it occurs.

The similarities do not exist in the solid phase temperature only, but also they can be witnessed in gas phase temperatures where except the time of the occurrence of peak, considerable agreement exists in the variation of temperatures with fractional axial distance. Fig. 4.61 shows the results from the N-Tanks in series approach and Fig. 4.62 shows the results from Von Meien and Mitchell (2002).

The water activity of air/solid has been defined as the ratio of equilibrated partial vapor pressures of water present in solid/gas and pure water. The plots for water activity of the gas/solid phase has been evaluated at the prevailing temperatures and water contents of gas/solid phase with the help of constitutive relationships 3.69 and 3.70 and thereafter compared with those obtained by Von Meien and Mitchell (2002). It can be clearly seen that except of the time of occurrence of the maximal temperature of the solids at the outlet (4.5 hrs. for N-Tanks in Series and 12 hrs. for Von Meien and Mitchell (2002)), there is a considerable agreement between the two studies as shown by figures 4.63 and 4.64 for water activity of solid phase ; and 4.65 and 4.66 for water activity of gas phase.

The probable reason for the discrepancy in time can be understood from the plots of biomass concentration vs. time at the mid-height of the packed bed obtained for N-Tanks in Series approach (Fig.4.67) and by Von Meien and Mitchell (Fig. 4.68). The biomass concentration at 6 hrs. is 0.088 kg biomass/kg solid, whereas the biomass concentration of 0.09 kg biomass/ kg

solid is attained in about 20 hrs. in Von Meien and Mitchell's study. Similarly on observing the profiles for water content of the solids in the packed bed it can be clearly seen that in the N-Tanks in Series approach at mid-height the bed attains a solid phase water content of around 0.15 kg water/kg dry solid in 6 hours in N-Tanks in Series (Fig. 4.69) and in 20 hrs. in Von Meien and Mitchell's study (Fig. 4.70). Hence in the case of N-Tanks in Series, the growth of biomass occurs at a faster rate which has accelerated the times for all variables.

On further examination of the cause, it was found that the units in which the mass transfer coefficient K_a was $\text{kg H}_2\text{O} / \text{s m}^3$. In the equations wherever K_a has been used, namely 3.76, 3.77 and 3.79, the consistency of units shall be established only if the values of K_a had been as

$$\frac{\text{kg H}_2\text{O}}{\text{m}^3 \text{ s} \left(\frac{\text{kg water}}{\text{kg total dry solids}} \right)}$$

This change could not be introduced in the model equations as such, since the values of K_a had been determined with the help of a correlation 3.71.

$$K_a = (a_1 + a_2 [T_g + 273]) \Phi_s - a_3 + a_4 (T_g + 273) \quad (3.71)$$

where $a_1 = 7.304$, $a_2 = 1.77 \times 10^{-2}$, $a_3 = 2.202$, $a_4 = -6.18 \times 10^{-3}$

The correlation has been taken from Von Meien and Mitchell (2002) which in turn quotes Mancini (1996), "Transferência de massa em secadores de grãos", PhD Thesis, Universidade Federal de Rio de Janeiro, Rio de Janeiro, Brazil as its source.

An internet publication of Mancini et al., "Transferência de massa na secagem de milho em secadores de camada espessa em leitos fixo e deslizante" is in Portuguese but cites the correlation similar to the above to exist in the form:-

$$k_s a = \alpha Y_s - \beta \quad (4.2)$$

where $\alpha = a_1 - a_2 T_g$ (4.3 and 4.4)
 $\beta = b_1 - b_2 T_g$

$$a_1 = 7.31 \text{ kg/m}^3 \text{ s}$$

$$a_2 = -1.77 \times 10^{-2} \text{ kg/m}^3 \text{ s K}$$

$$b_1 = 2.20 \text{ kg/m}^3 \text{ s}$$

$$b_2 = -6.18 \times 10^{-3} \text{ kg/m}^3 \text{ s K}$$

If the above relationships are incorporated into the computer program then the results as shown in Fig. 4.71-4.76 are obtained. In these results the extent of drying is very less, which is inconsistent, where the packed bed should lose considerable moisture when it reaches temperatures above 50°C. Hence the correlation for mass transfer coefficient needs to be looked into greater detail before going for a solution of the two-phase model. It is possible that some details regarding the usage of the correlation are not mentioned in the paper of Von Meien and Mitchell (2002).

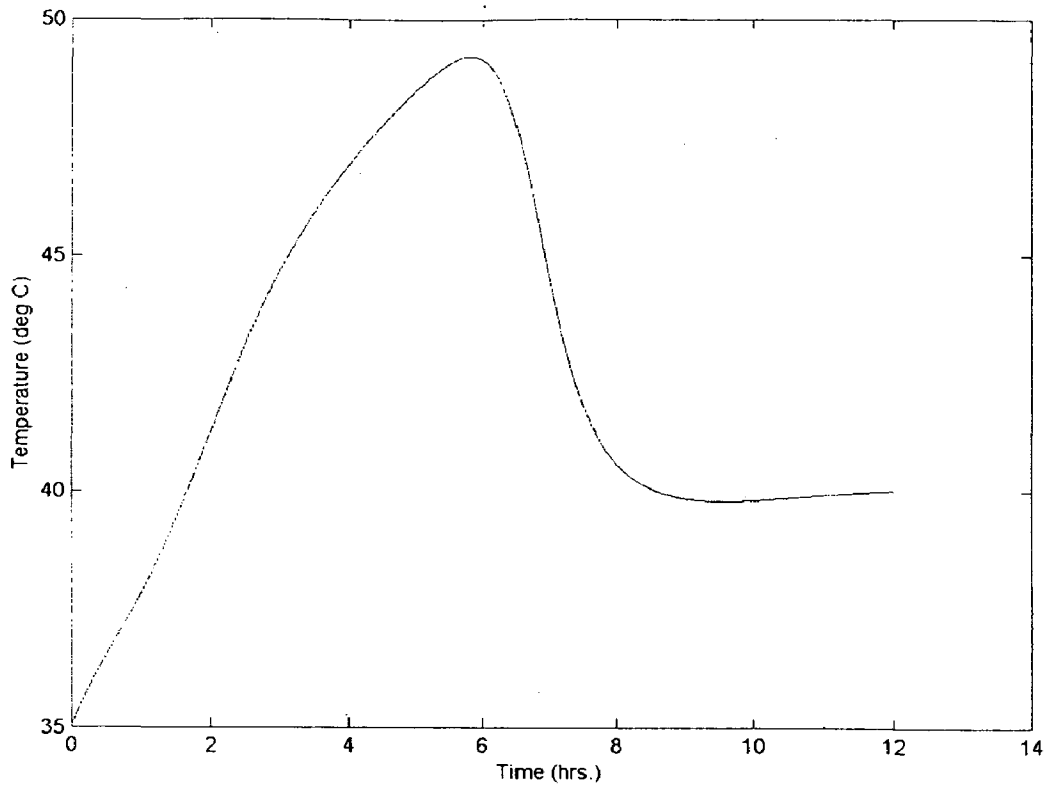


Figure 4.41 Plot of temperature of solid phase vs. time for $N = 1$

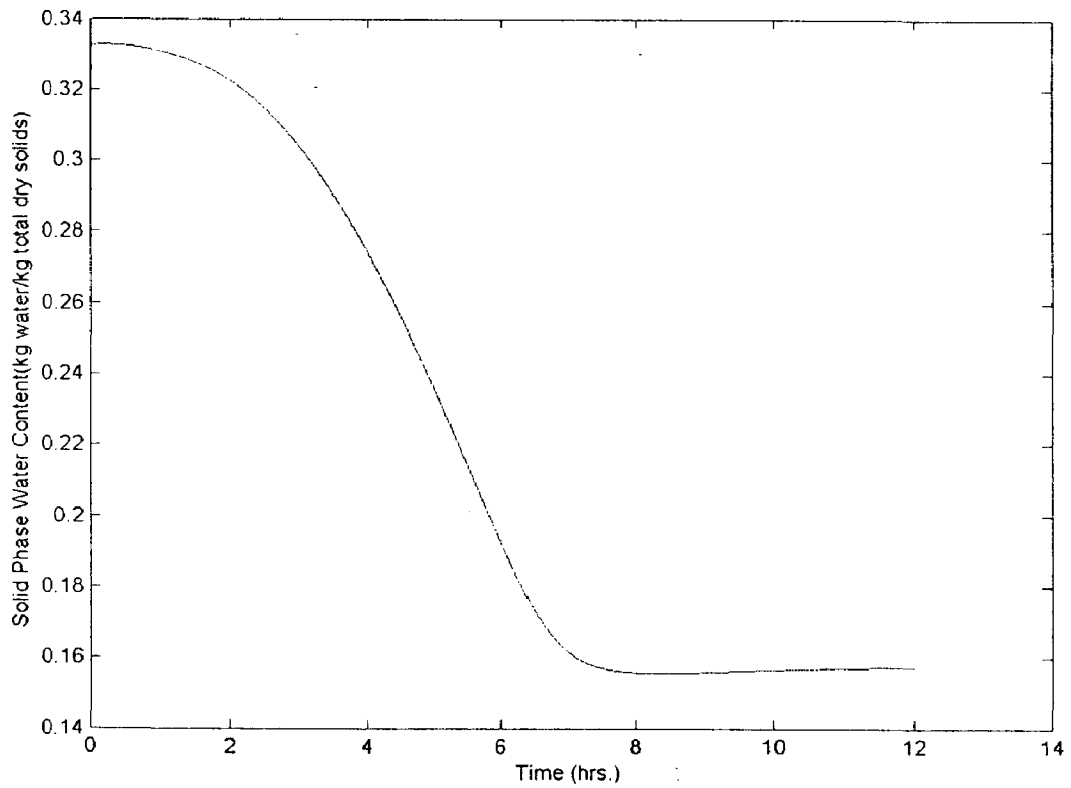


Figure 4.42 Plot of water content of solid phase vs. time for $N = 1$

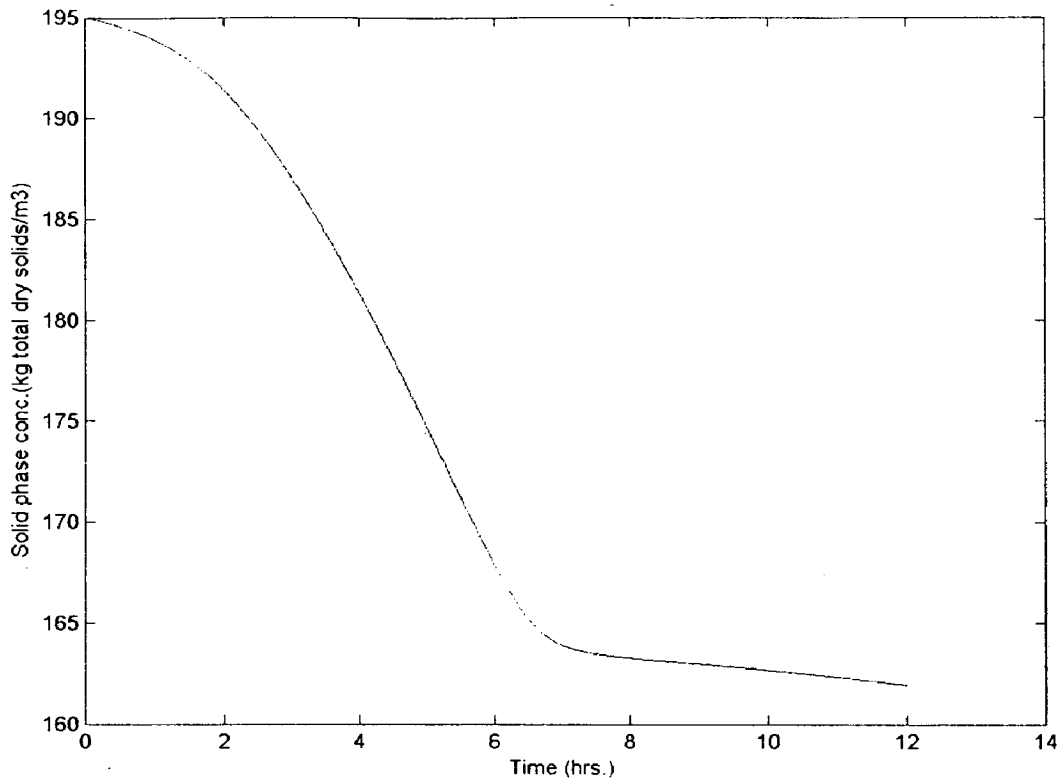


Figure 4.43 Plot of concentration of total dry solids vs. time for $N = 1$

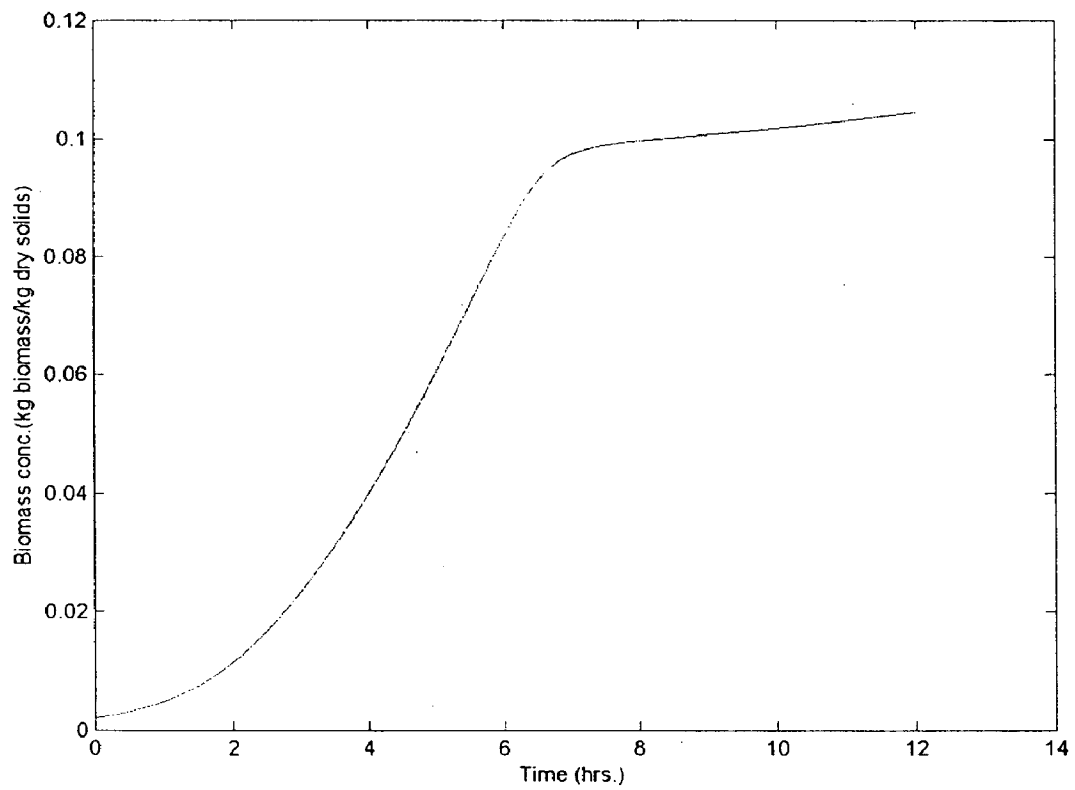


Figure 4.44 Plot of biomass concentration vs. time for $N = 1$

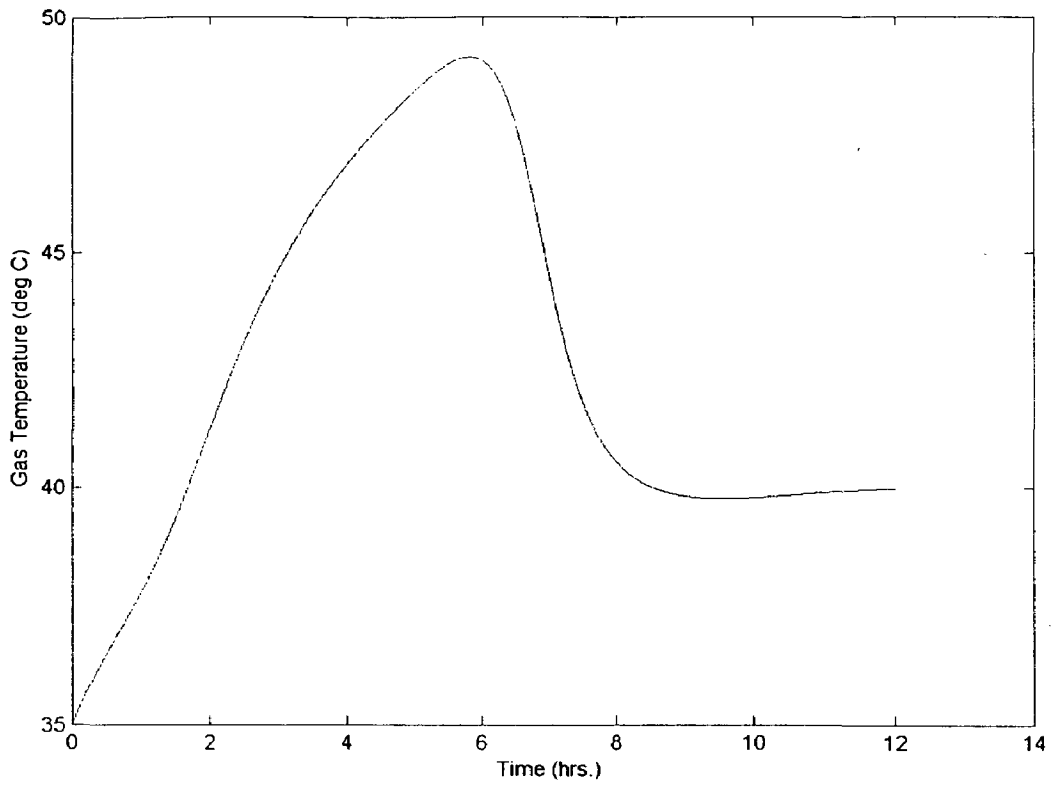


Figure 4.45 Plot of air(gas phase) temperature vs. time for N = 1

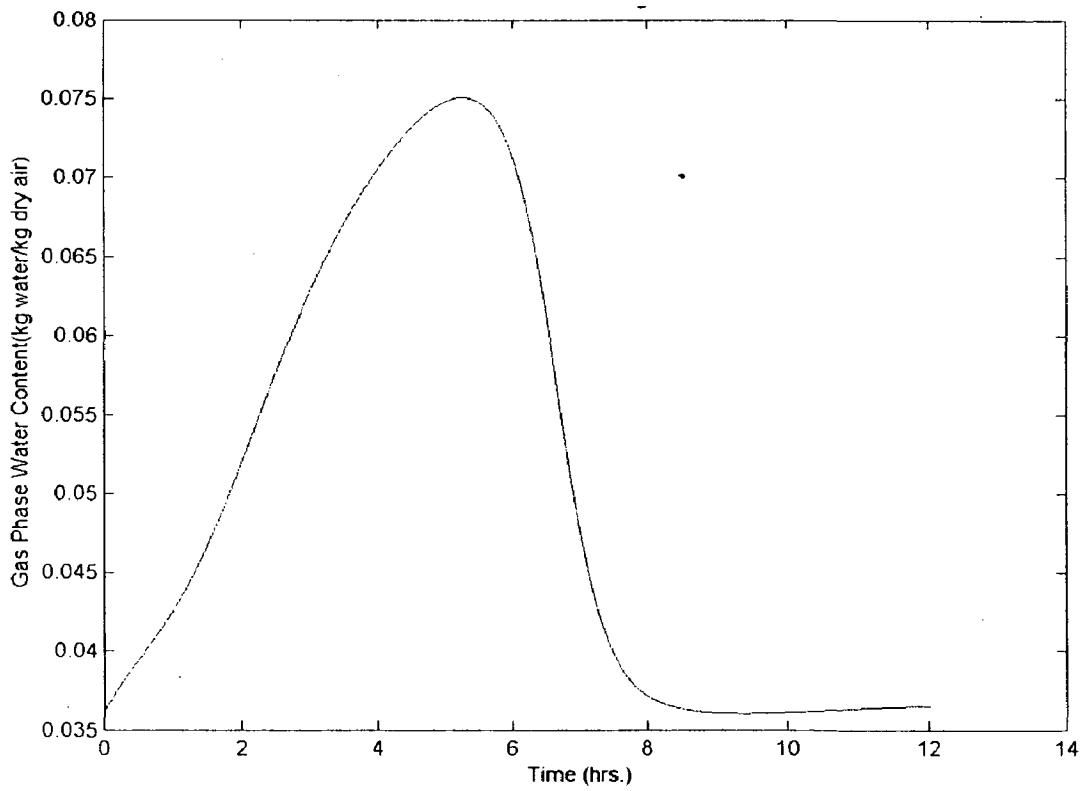


Figure 4.46 Plot of moisture content of air(gas phase) vs. time for N = 1

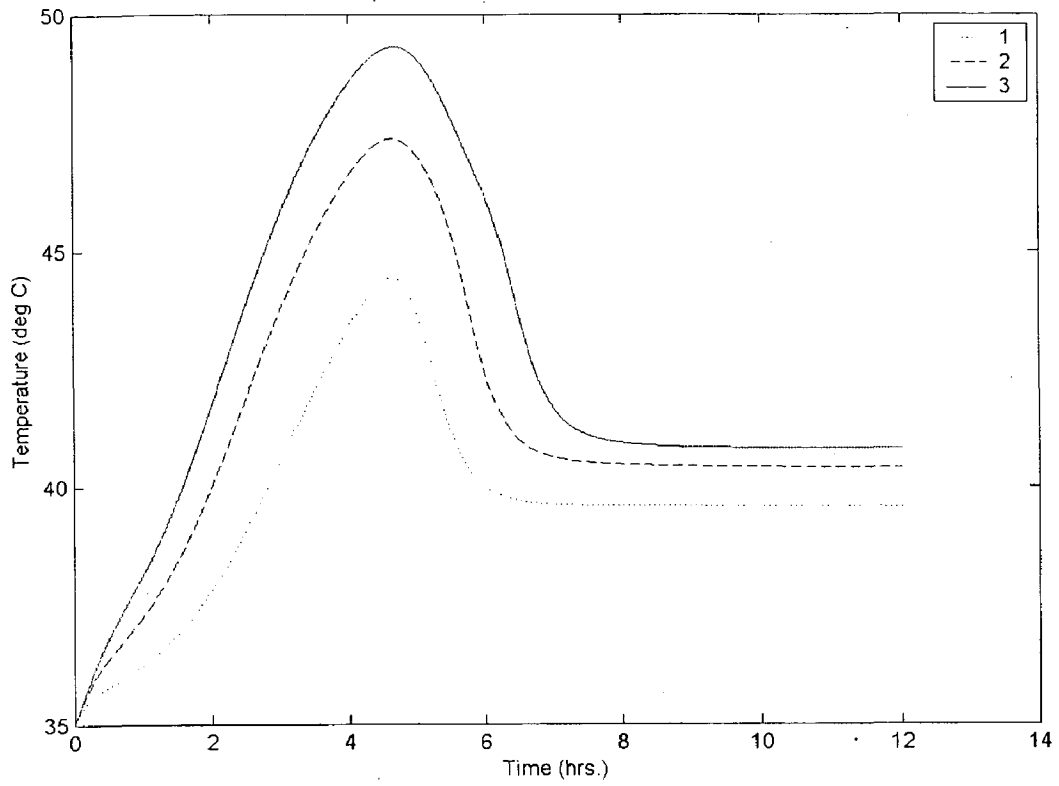


Figure 4.47 Plot of temperature of solid phase vs. time for N = 3

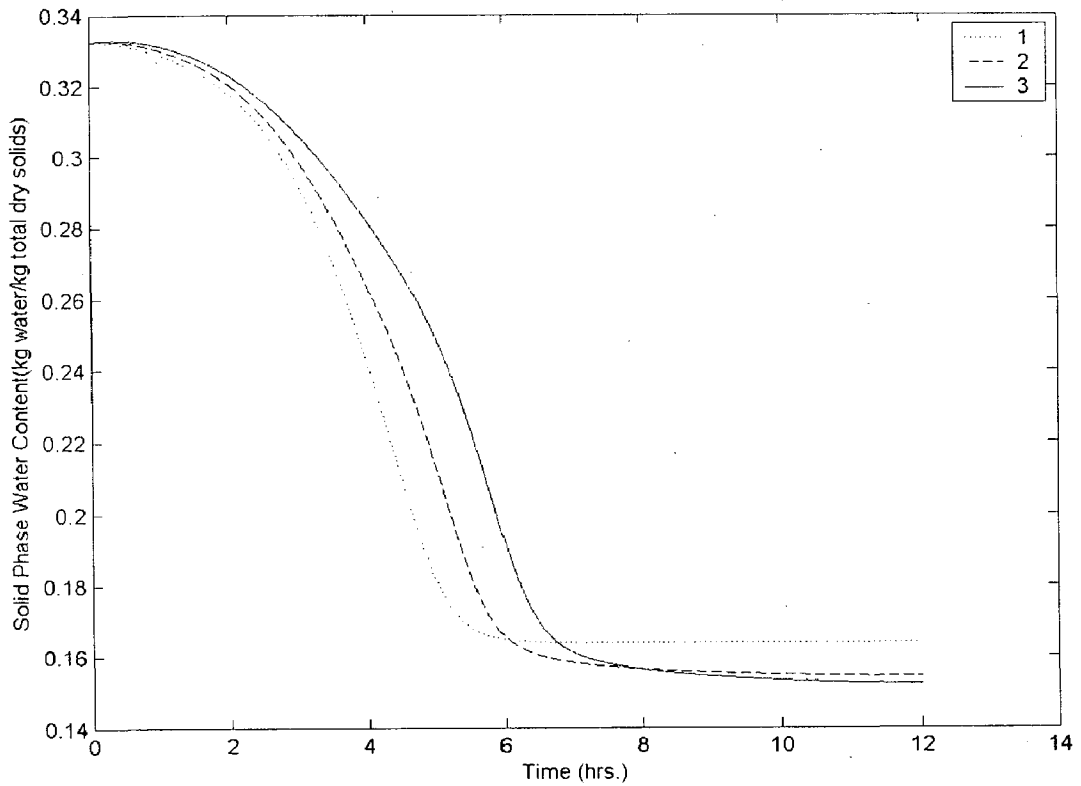


Figure 4.48 Plot of water content of solid phase vs. time for N = 3

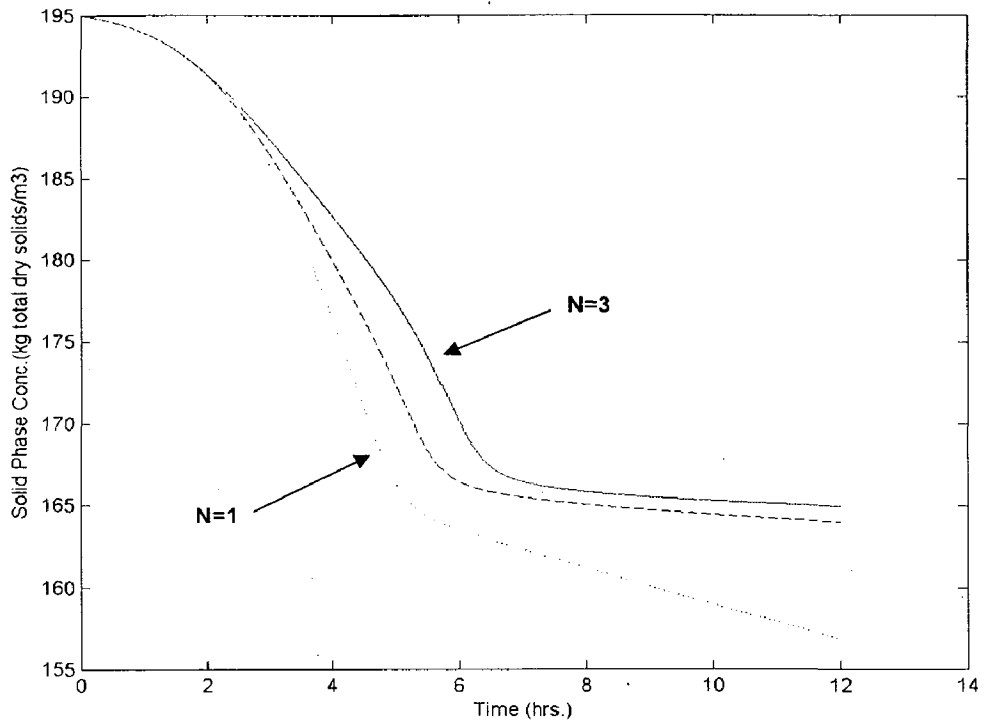


Figure 4.49 Plot of solid phase concentration vs. time for $N = 3$

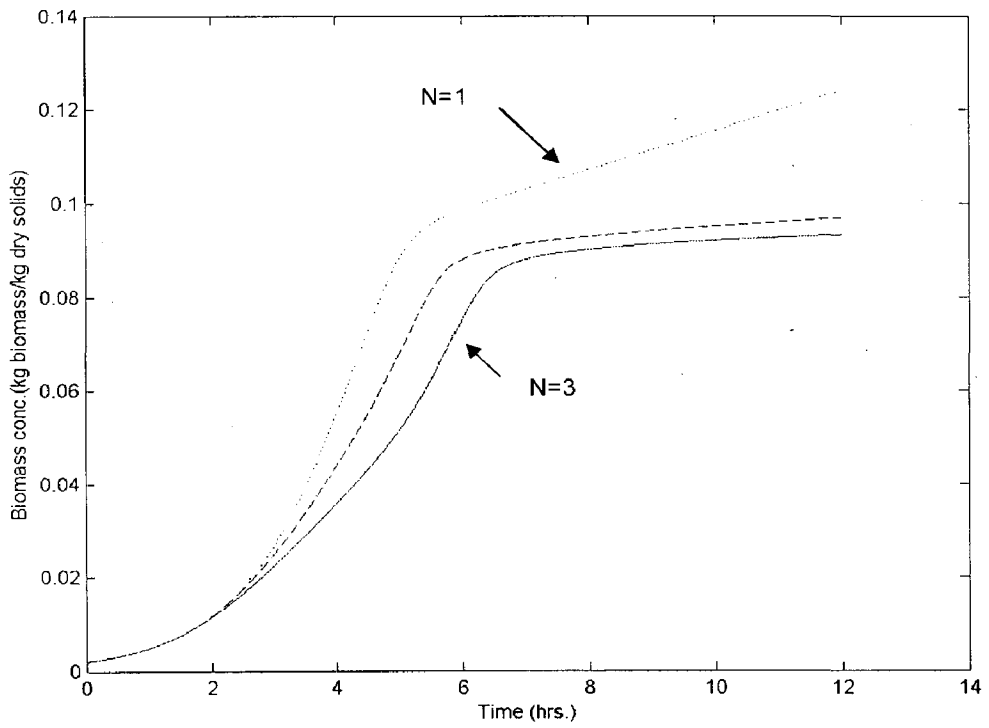


Figure 4.50 Plot of biomass concentration vs. time for $N = 3$

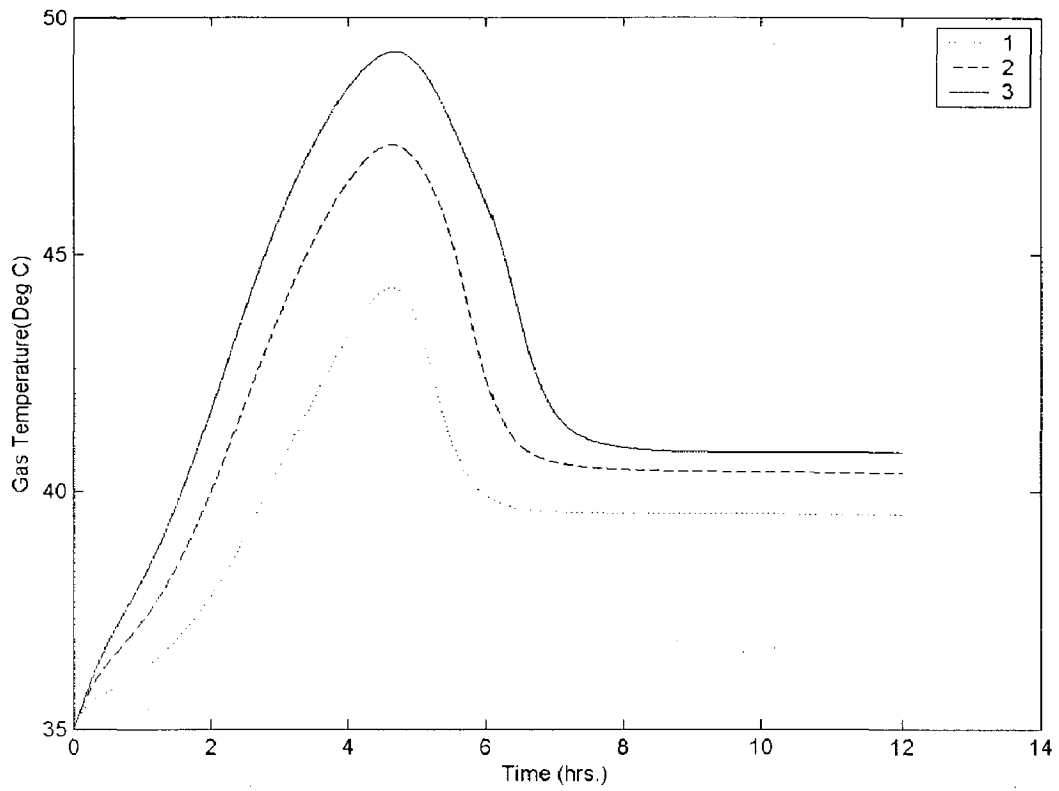


Figure 4.51 Plot of gas phase (air) temperature vs. time for $N = 3$

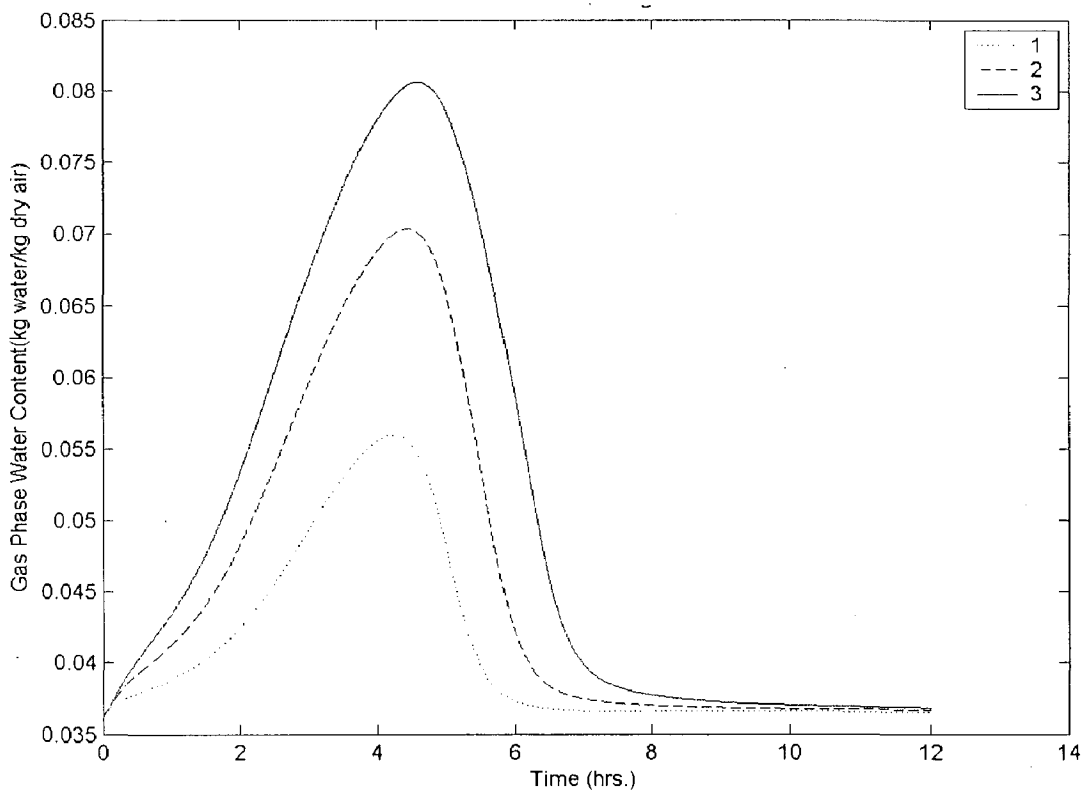


Figure 4.52 Plot of gas phase (air) water content vs. time for $N = 3$

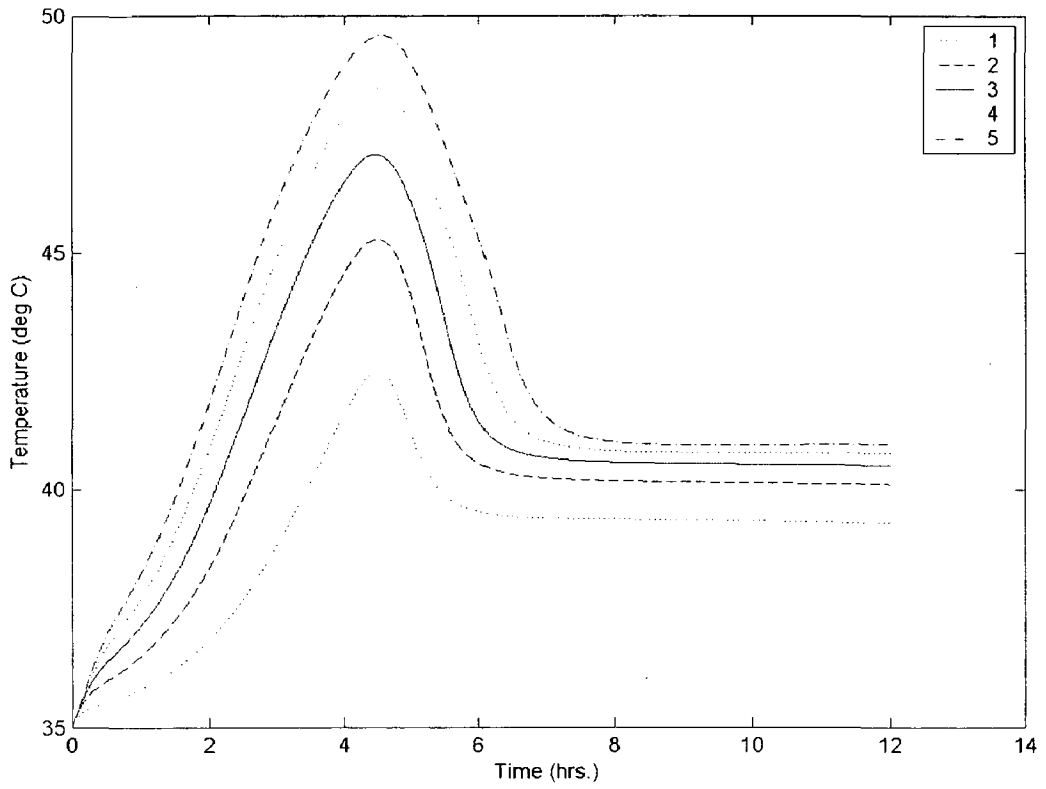


Figure 4.53 Plot of temperature of solid phase vs. time for $N = 5$

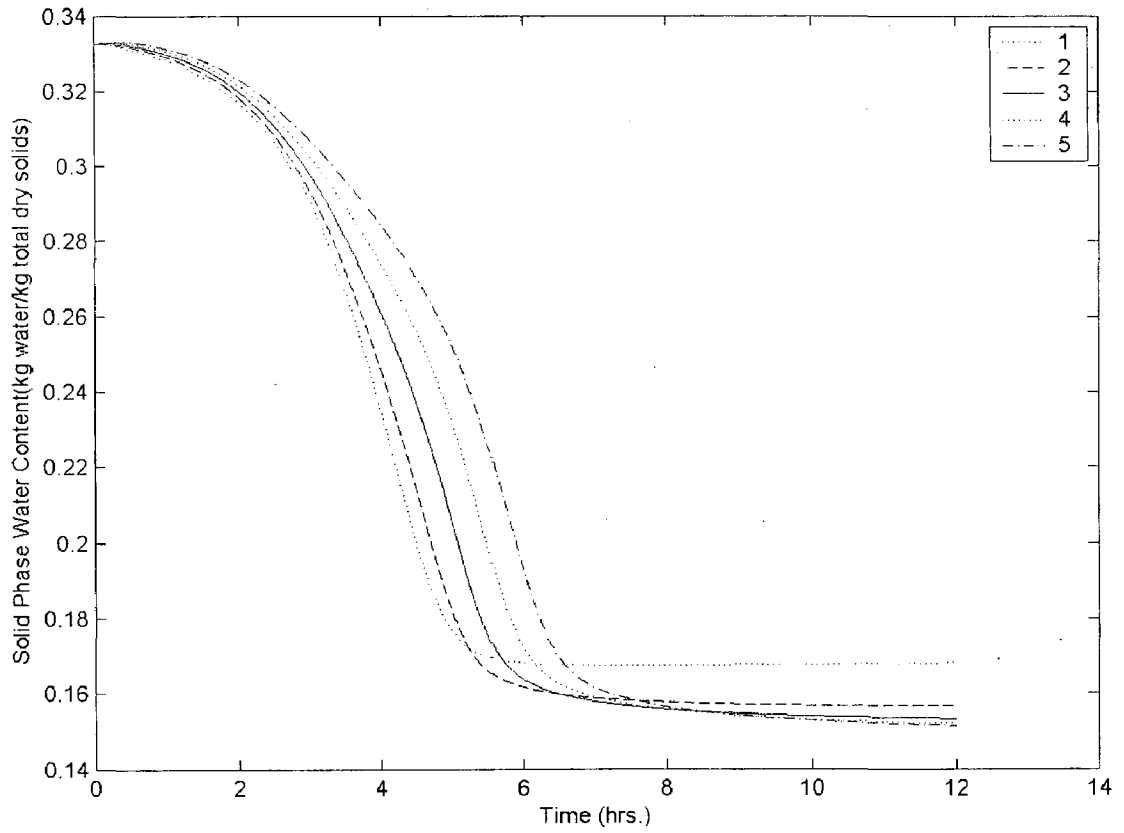


Figure 4.54 Plot of water content of solid phase vs. time for $N = 5$

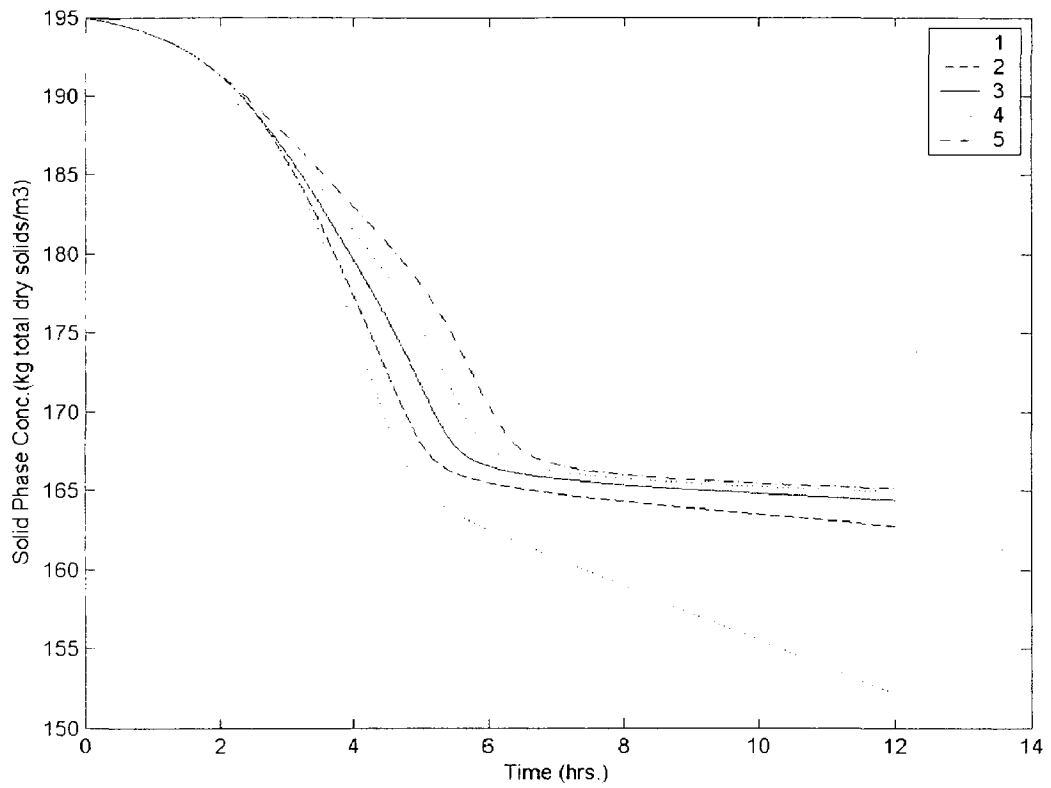


Figure 4.55 Plot of solid phase concentration vs. time for N = 5

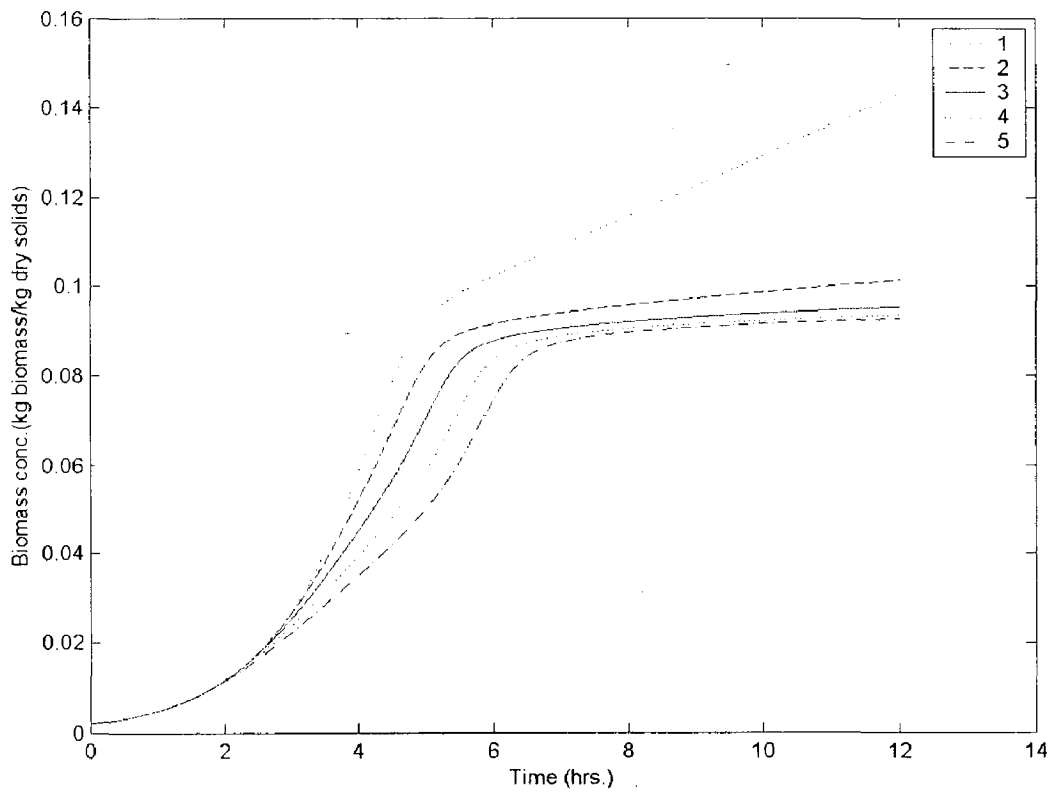


Figure 4.56 Plot of biomass concentration vs. time for N = 5

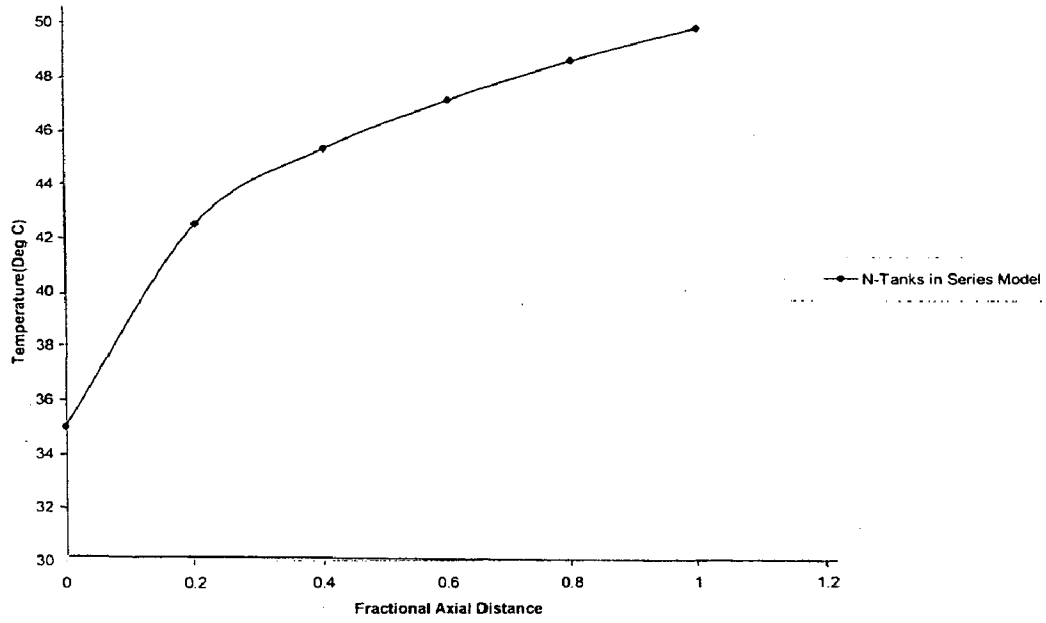


Figure 4.59 Plot of temperature of solid phase vs. axial distance at $t=4.5$ hours as determined by N-Tanks in Series model at which the solids temperature at the outlet is near maximal.

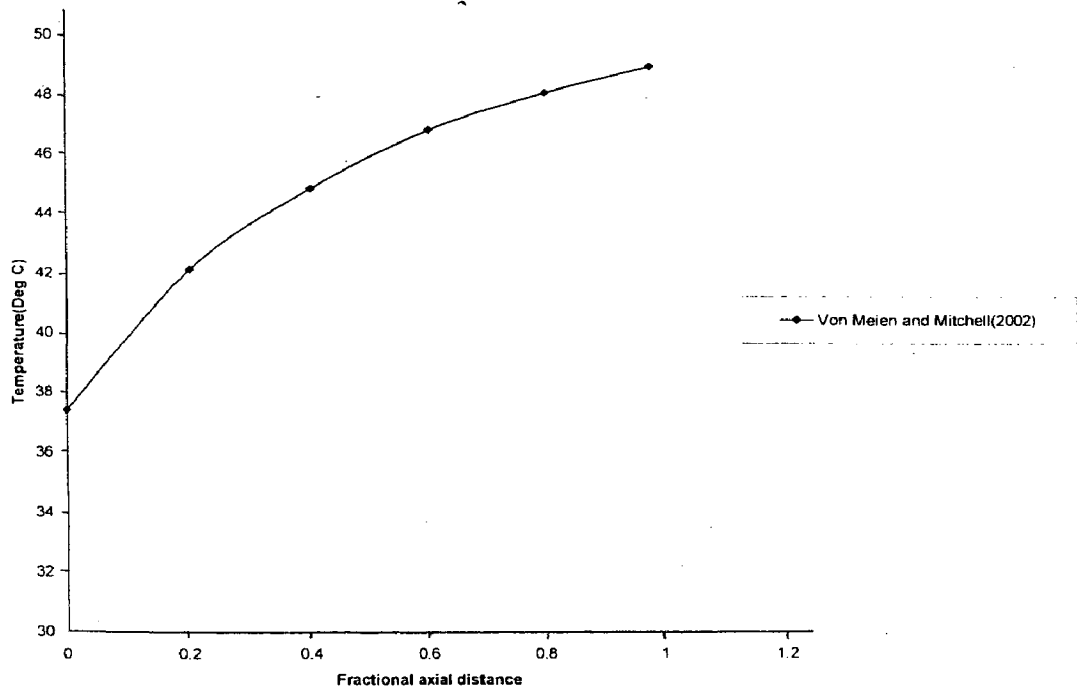


Figure 4.60 Plot of temperature of solid phase vs. axial distance at $t=12$ hours as determined by Von Meien and Mitchell (2002) at which the solids temperature at the outlet is near maximal.

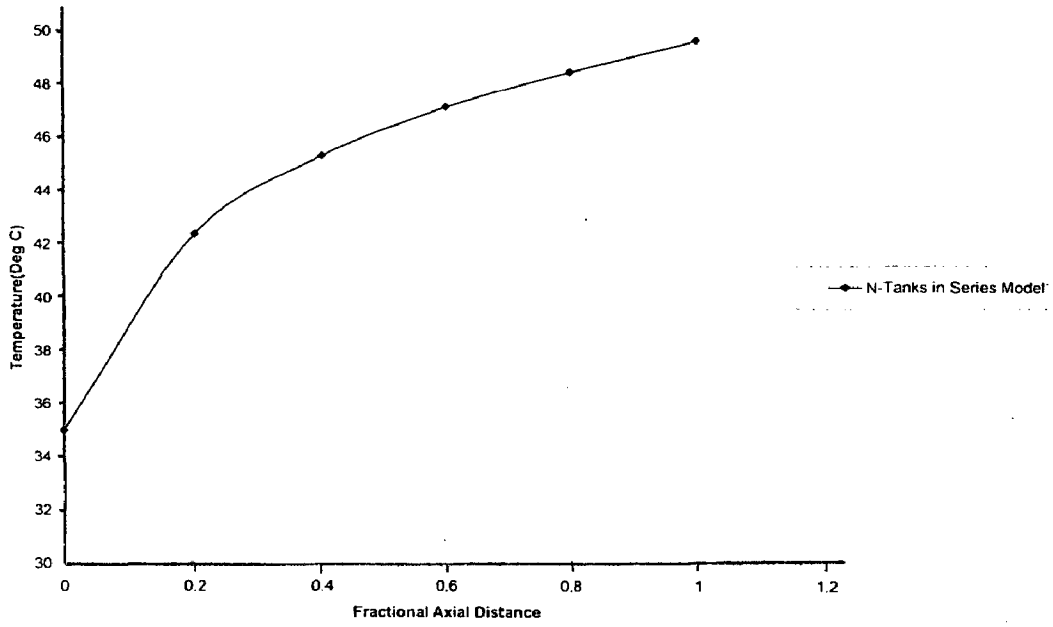


Figure 4.61 Plot of temperature of gas phase vs. axial distance at $t = 4.5$ hours as determined by N-Tanks in Series model at which the solids temperature at the outlet is near maximal.

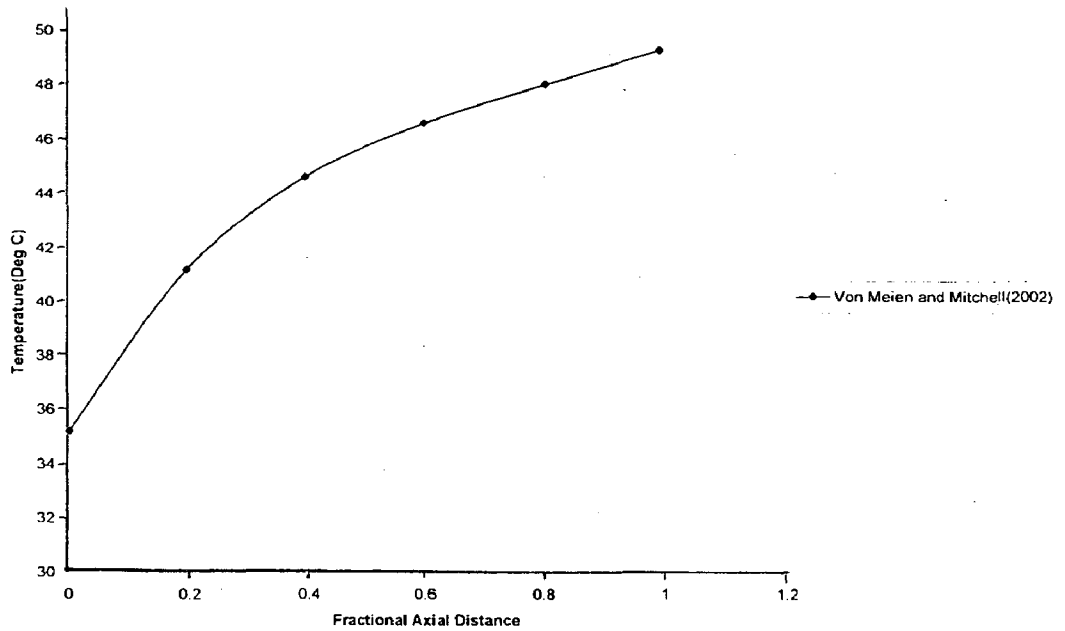


Figure 4.62 Plot of temperature of gas phase vs. axial distance at $t = 12$ hours as determined by Von Meien and Mitchell (2002) at which the solids temperature at the outlet is near maximal.

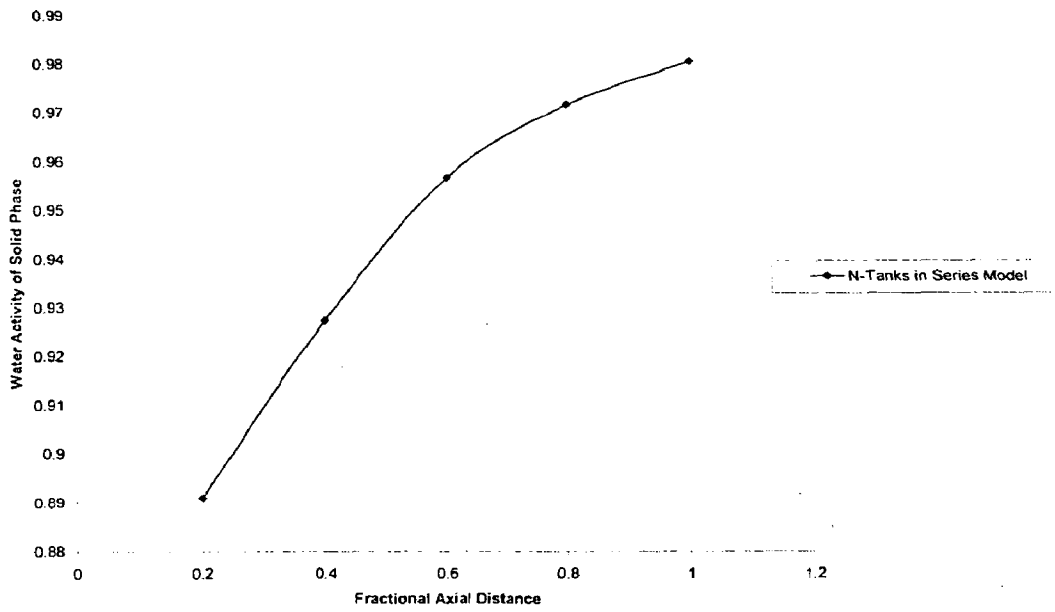


Figure 4.63 Plot of water activity of solid phase vs. axial distance at $t = 4.5$ hours as determined by N-Tanks in Series model at which the solids temperature at the outlet is near maximal.

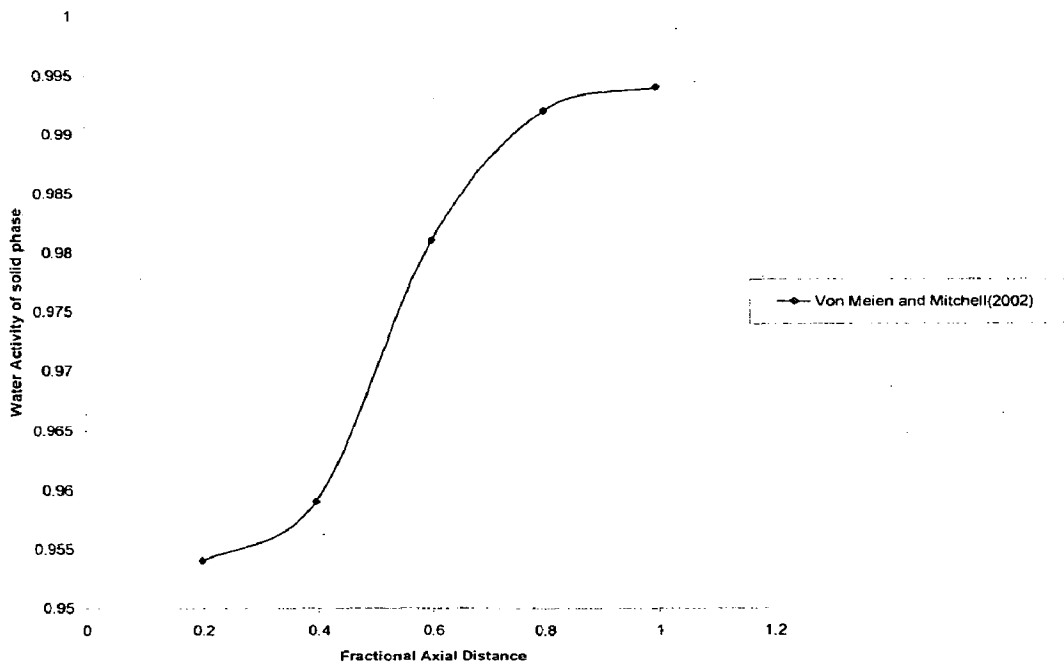


Figure 4.64 Plot of water activity of solid phase vs. axial distance at $t = 12$ hours as determined by Von Meien and Mitchell(2002) at which the solids temperature at the outlet is near maximal.

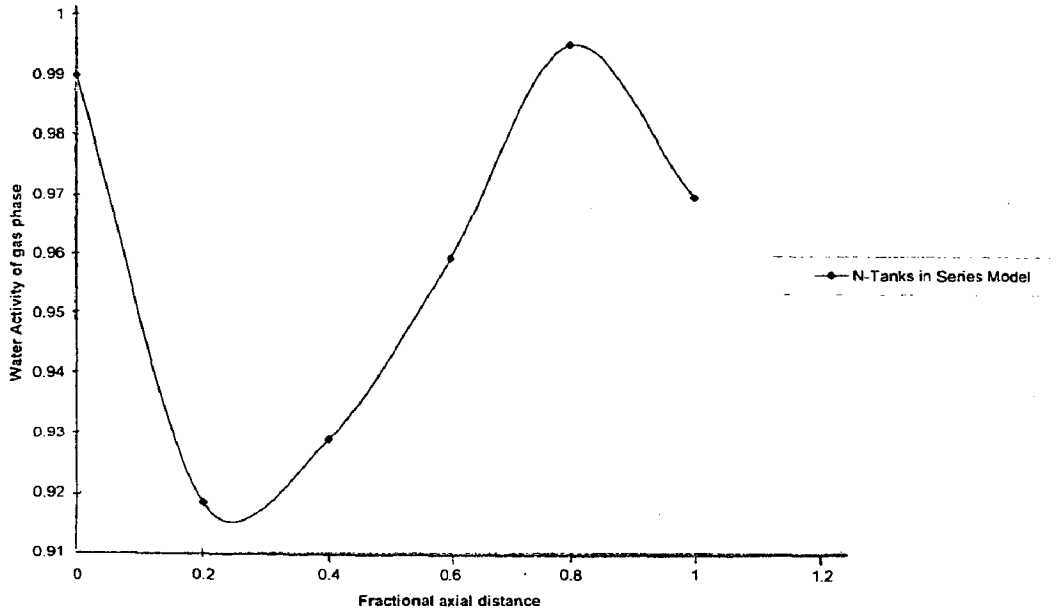


Figure 4.65 Plot of water activity of gas phase vs. axial distance at $t=4.5$ hours as determined by N-Tanks in Series model at which the solids temperature at the outlet is near maximal.

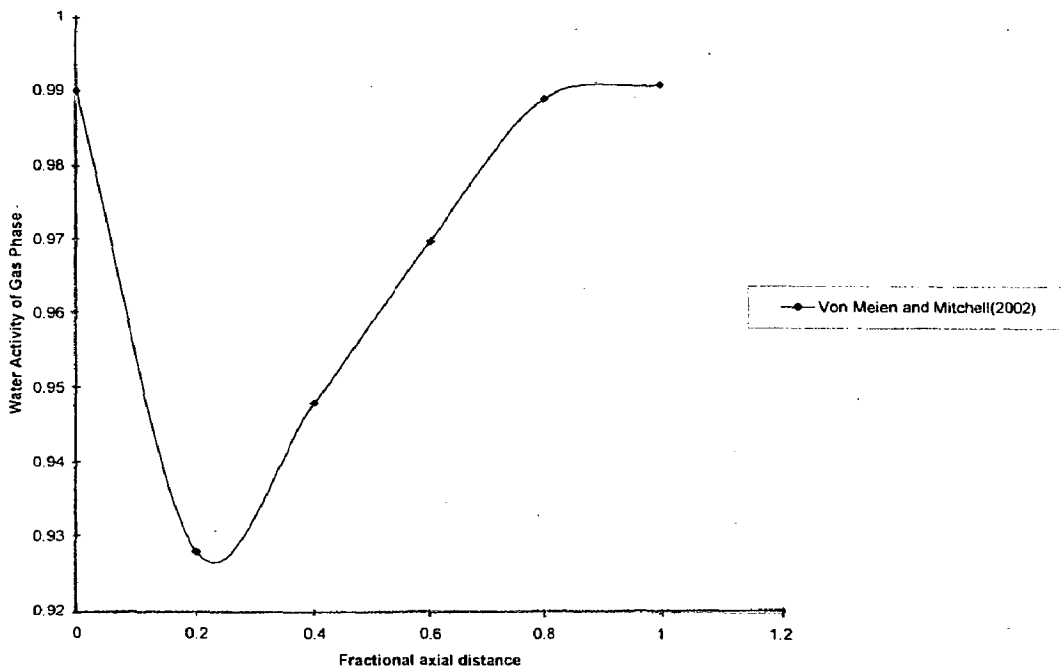


Figure 4.66 Plot of water activity of gas phase vs. axial distance at $t=12$ hours as determined by Von Meien and Mitchell (2002) at which the solids temperature at the outlet is near maximal.

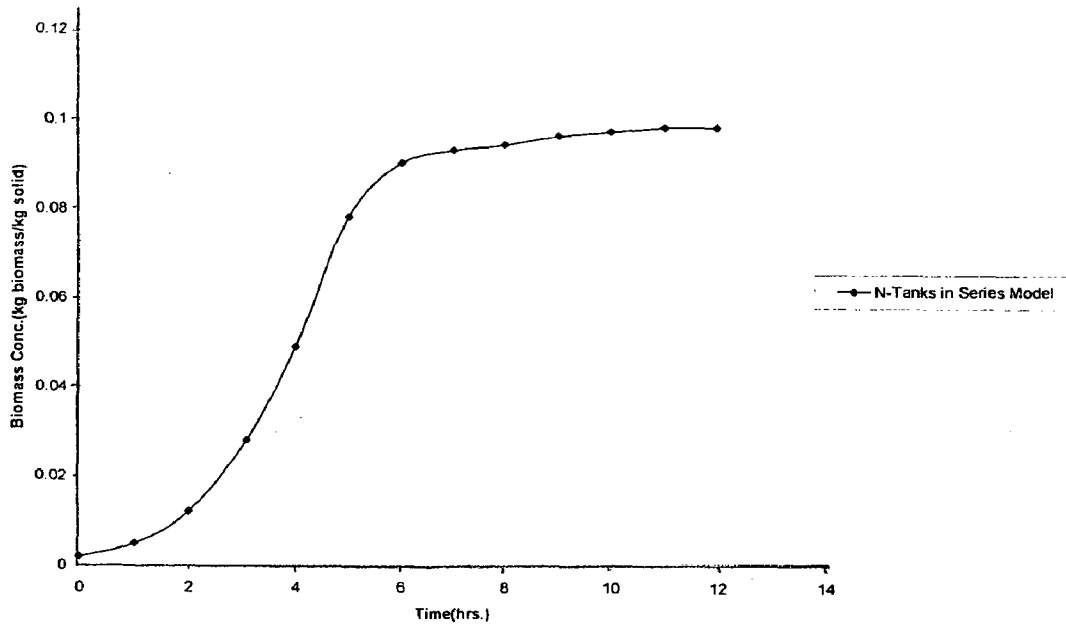


Figure 4.67 Plot of Biomass Concentration vs. time at the mid-height of bed as determined by N-Tanks in Series model.

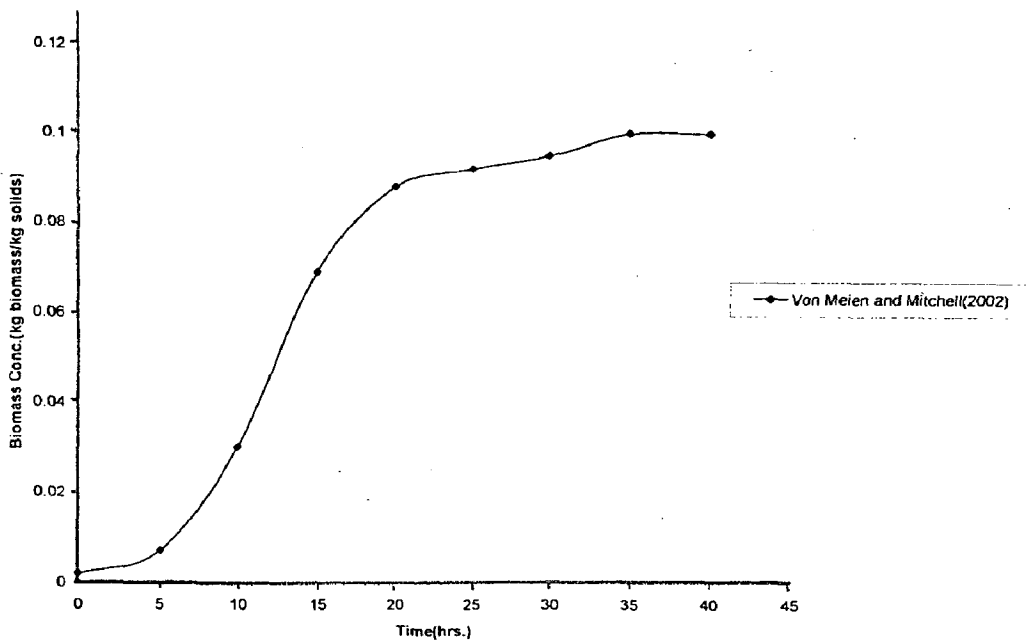


Figure 4.68 Plot of Biomass Concentration vs. time at the mid-height of bed as determined by Von Meien and Mitchell(2002).

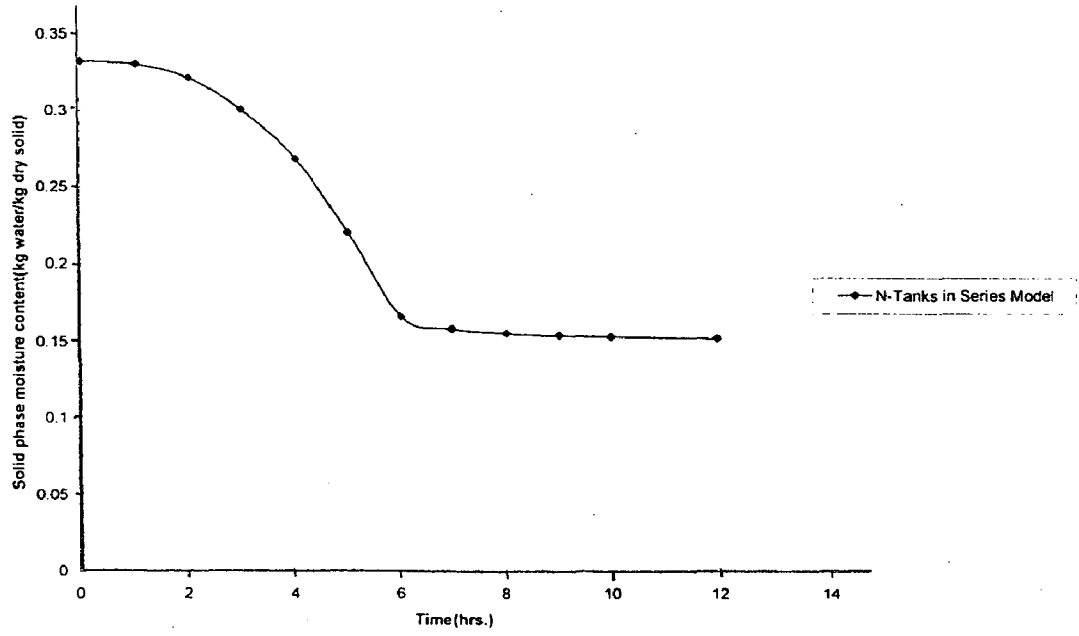


Figure 4.69 Plot of Water Content of Solid Phase vs. time at the mid-height of bed as determined by N-Tanks in Series model.

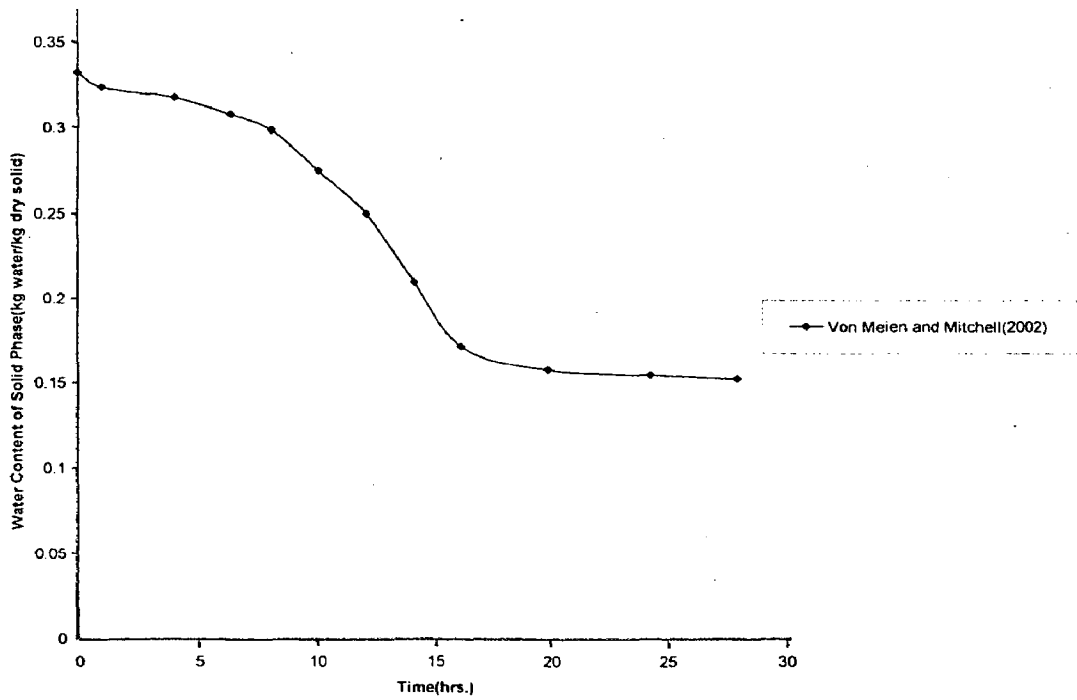


Figure 4.70 Plot of Water Content of Solid Phase vs. time at the mid-height of bed as determined by Von Meien and Mitchell(2002).

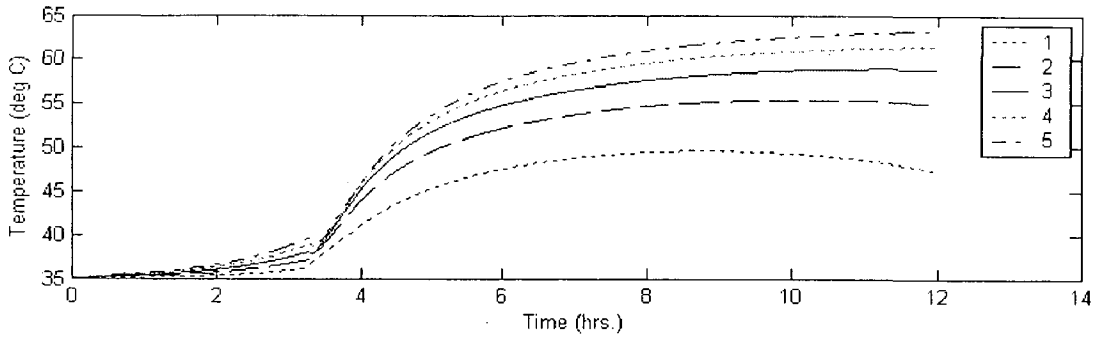


Figure 4.71 Plot of Temperature of solid phase vs. time ,for the correlation cited in Mancini et al. as determined by N-Tanks in Series model.

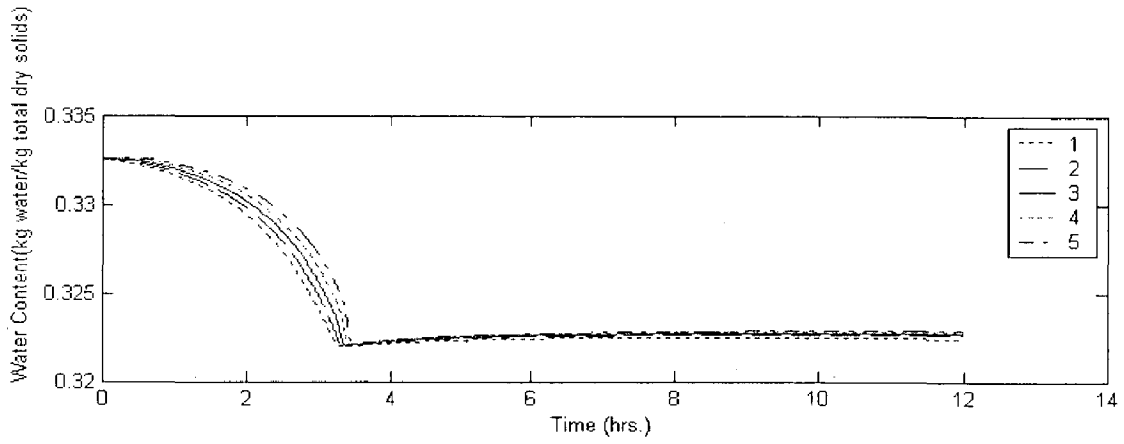


Figure 4.72 Plot of Water Content of solid phase vs. time, for the correlation cited in Mancini et al. as determined by N-Tanks in Series model.

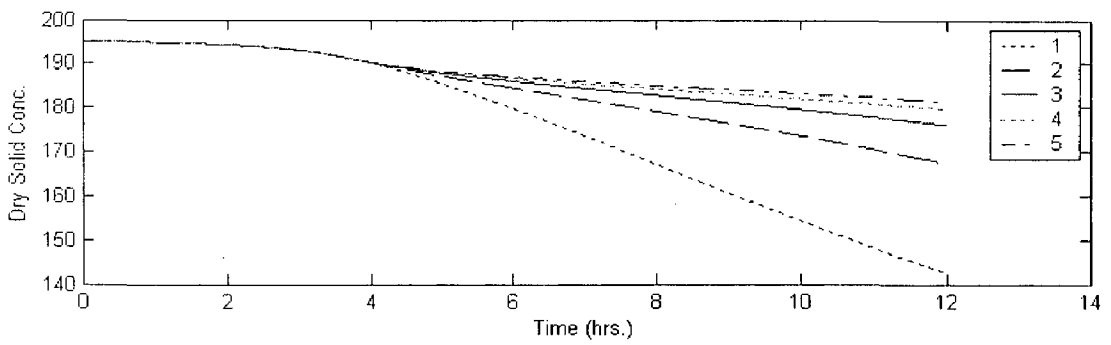


Figure 4.73 Plot of solid phase concentration vs. time, for the correlation cited in Mancini et al. as determined by N-Tanks in Series model.

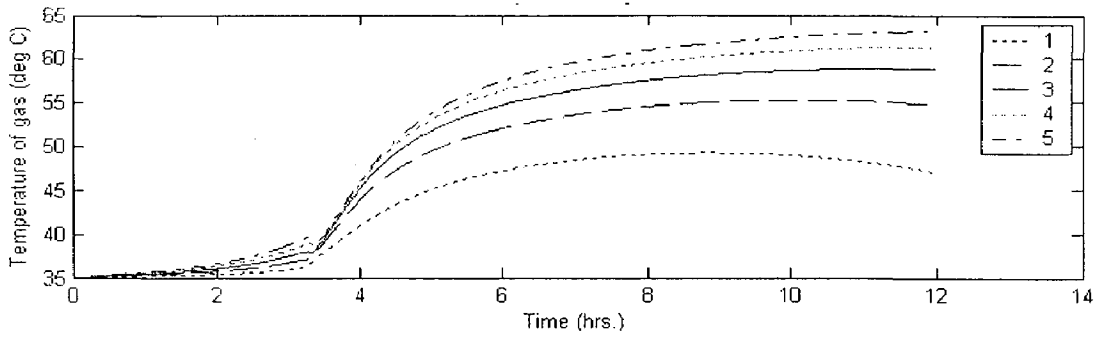


Figure 4.74 Plot of temperature of gaseous phase vs. time, for the correlation cited in Mancini et al. as determined by N-Tanks in Series model.

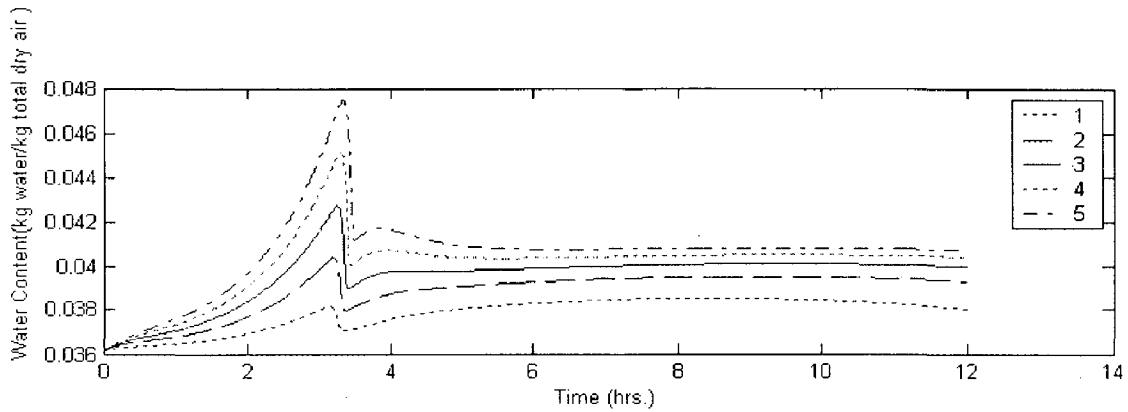


Figure 4.75 Plot of moisture content of gaseous phase vs. time, for the correlation cited in Mancini et al. as determined by N-Tanks in Series model

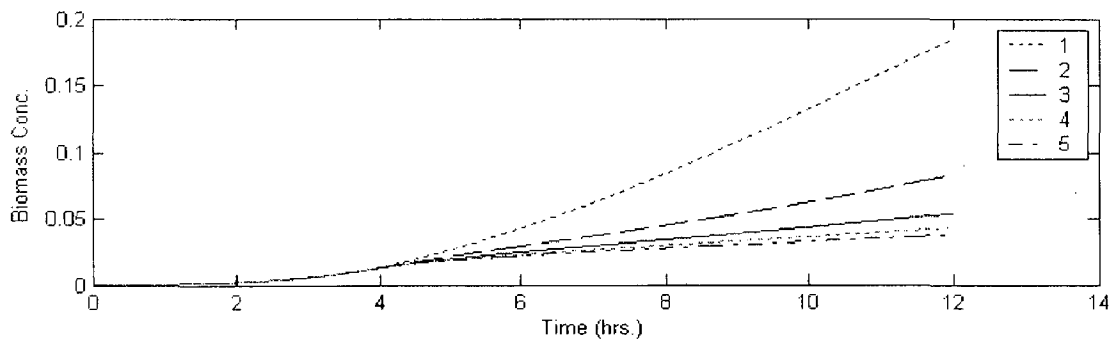


Figure 4.76 Plot of Biomass Concentration vs. time, for the correlation cited in Mancini et al. as determined by N-Tanks in Series model

4.3 CONCLUDING REMARKS

The results from the solution of mathematical models developed by employing the N-Tanks in Series approach for the Packed Bed Solid State Fermentation Bioreactor considering pseudohomogeneous and two phase systems demonstrate the potential for its wide scale of application. The model has been validated with the experimental data and has found to be less computationally complex to implement without loss in accuracy.

CONCLUSIONS AND RECOMMENDATIONS

5.1 CONCLUSIONS

The development of the mathematical model of the Packed Bed Solid State Fermentation Packed Bed Bioreactor by the N-Tanks in Series approach envisaged two fundamental aims:

- i) To attempt and extend the application of Chemical Reaction Engineering concepts in the formulation of a mathematical model of a biological system which would not only ease the complexities associated with computation involved in the determination of profiles of variables like temperature ,concentration ,water content with length and time.
- ii) The developed model should help in providing insights which could be useful in providing basic design estimates.

In this thesis, we have developed the mathematical model for two different cases, namely if the solid phase and gaseous phase in the packed bed are modelled as a pseudohomogeneous phase; and if the two phases are treated as distinct phases.

5.1.1 Pseudohomogeneous Phase Model developed by N-Tanks in Series approach

A mathematical model employing the N-Tanks in series approach had been developed which considered the production of protease enzyme by two different species namely *Aspergillus niger* and *Penicillium fellutanum*.

The model constituted an energy balance, equations describing the growth of biomass by logistic growth kinetics, production of solid substrate active enzyme and denaturation of enzyme. The model equations were solved by a program incorporating the Runge-Kutta-Fehlberg algorithm in MATLAB. The profiles of temperature, solid substrate active enzyme content

and biomass versus time for both the biological species were found out. The variations of the aforesaid variables with axial distance (height) were also determined which could prove to be of immense importance in aiding the determination of the requisite height of the bioreactor.

The model was validated with the help of the experimental data of Ghildyal et al. (1994) and a good agreement was found to exist between the two. On comparison of the validation of N-Tanks in Series approach with that done employing Orthogonal Collocation by Sangsurasak and Mitchell (1998), it was found that the complexities in the computational procedure can be considerably reduced if the N-Tanks in Series approach was employed.

5.1.2 Two-Phase Model developed by N-Tanks in Series approach

The growth of *Aspergillus niger* on Corn was modeled as a two-phase system by the N-Tanks in series approach. Von Meien and Mitchell (2002) had described the above system as a set of partial differential equations. The N-Tanks in series approach was employed to convert the partial differential equations to ordinary differential equations. The computational experience regarding the system of equations was that they constituted a stiff system of Ordinary Differential Equations. To gain insights in the process a program employing the Runge Kutta Fourth Order Method in MATLAB was developed which constituted the model equations employing energy balance over the solid and gaseous phases, mass balance of water over the solid and gaseous phases in the packed bed ; and solid and biomass concentration. The solution of the model equations gave the profile of temperatures and water contents of solid and gaseous phases; substrate and biomass concentrations with time.

On comparison with the profiles as those obtained by Von Meien and Mitchell(2002), it was found with the N-Tanks in Series model ,identical profiles of temperature, water content ,water activity in the gas and solid phases were obtained but at different times. The reason of the discrepancy has to be looked upon. It may be attributed to the evaluation and the form of correlation of mass transfer coefficient, cited by Von Meien and Mitchell (2002).

In spite of the discrepancy, the occurrence of identical profiles in magnitude proves the potential of the usage of N-Tanks in Series Model to systems modeled as two phase system.

5.1.3 Final Remarks

The modeling approach proposed and envisaged in the thesis for Packed Bed Solid State Fermentation Bioreactors can be successfully used not only for the analysis and design for systems modeled as a pseudohomogeneous or two phase, but also aid in the simplification of computational complexities which have been involved in solving models involving partial differential equations which have been proposed till date.

5.2 RECOMMENDATIONS FOR FUTURE WORK

The research work in the present work could be extended further by executing the set of model equations developed for routines developed especially for stiff differential equation solvers like DASSL, ode15s routine in MATLAB etc. An analysis of stiffness ratio could be applied to the model equations of two-phase system could be done to identify possibilities for easing out computation.

Any further improvement or development of the proposed model should incorporate the proper description of the mass transfer process of water in both the solid and gaseous phases through correlations which should be consistent in their units. It is imperative that more experimental work be carried out, which hampers broader use of mathematical modeling as also suggested by Lenz et al.(2004).

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APPENDIX A: RUNGE-KUTTA-FEHLBERG FORMULAE FOR A SYSTEM OF ORDINARY DIFFERENTIAL EQUATIONS

Consider the following initial value problem which consists of the following equations:

$$\frac{d\vec{y}}{dt} = \vec{f}(t, \vec{y}) \quad (\text{A.1a})$$

$$\vec{y} = \vec{y}_0 \text{ at } t = t_0 \quad (\text{A.1b})$$

where

$$\vec{y}^T = [y_1 \ y_2 \ \dots \ y_n] \quad (\text{A.2})$$

$$\vec{y}_0^T = [y_{10} \ y_{20} \ \dots \ y_{n0}] \quad (\text{A.3})$$

$$\vec{y}^T(t, \vec{y}) = [f_1(t, \vec{y}) \ f_2(t, \vec{y}) \ \dots \ f_n(t, \vec{y})]$$

or
$$\vec{f}^T = [f_1 \ f_2 \ f_3 \ \dots \ f_n] \quad (\text{A.4})$$

The Runge –Kutta –Fehlberg fifth order formula is:-

$$\vec{y}_{k+1} = \vec{y}_k + \frac{16}{135} \vec{k}_1 + \frac{6656}{12825} \vec{k}_3 + \frac{28561}{564360} \vec{k}_4 - \frac{9}{50} \vec{k}_5 + \frac{2}{55} \vec{k}_6 \quad (\text{A.5})$$

where

$$\vec{k}_i^T = [\bar{k}_{1i} \ \bar{k}_{2i} \ \bar{k}_{3i} \ \dots \ \bar{k}_{ni}] \quad ; i=1 \text{ to } 4 \quad (\text{A.6})$$

Expressions for \bar{k}_1 to \bar{k}_6 are as follows:-

$$\begin{aligned}
\bar{k}_1 &= h\bar{f}[t_k, y_k] \\
\bar{k}_2 &= h\bar{f}\left[\left(t_k + \frac{1}{4}h\right), \left(y_k + \frac{1}{4}\bar{k}_1\right)\right] \\
\bar{k}_3 &= h\bar{f}\left[\left(t_k + \frac{3}{8}h\right), \left(y_k + \frac{3}{32}\bar{k}_1 + \frac{9}{32}\bar{k}_2\right)\right] \\
\bar{k}_4 &= h\bar{f}\left[\left(t_k + \frac{12}{13}h\right), \left(y_k + \frac{1932}{2197}\bar{k}_1 - \frac{7200}{2197}\bar{k}_2 + \frac{7296}{2197}\bar{k}_3\right)\right] \\
\bar{k}_5 &= h\bar{f}\left[\left(t_k + h\right), \left(y_k + \frac{439}{216}\bar{k}_1 - 8\bar{k}_2 + \frac{3680}{513}\bar{k}_3 - \frac{845}{4104}\bar{k}_4\right)\right] \\
\bar{k}_6 &= h\bar{f}\left[\left(t_k + \frac{1}{2}h\right), \left(y_k - \frac{8}{27}\bar{k}_1 + 2\bar{k}_2 - \frac{3544}{2565}\bar{k}_3 + \frac{1859}{4104}\bar{k}_4 - \frac{11}{40}\bar{k}_5\right)\right]
\end{aligned} \tag{A.7-A.12}$$

APPENDIX B: RUNGE-KUTTA FOURTH ORDER FORMULAE FOR A SYSTEM OF ORDINARY DIFFERENTIAL EQUATIONS

Consider the following initial value problem which consists of the following equations:

$$\frac{d\bar{y}}{dt} = \bar{f}(t, \bar{y}) \quad (\text{B.1a})$$

$$\bar{y} = \bar{y}_0 \text{ at } t = t_0 \quad (\text{B.1b})$$

where

$$\bar{y}^T = [y_1 \ y_2 \ \dots \ y_n] \quad (\text{B.2})$$

$$\bar{y}_0^T = [y_{10} \ y_{20} \ \dots \ y_{n0}] \quad (\text{B.3})$$

$$\bar{y}^T(t, \bar{y}) = [f_1(t, \bar{y}) \ f_2(t, \bar{y}) \ \dots \ f_n(t, \bar{y})] \quad \text{or}$$

$$\bar{f}^T = [f_1 \ f_2 \ f_3 \ \dots \ f_n] \quad (\text{B.4})$$

The fourth-order Runge Kutta Method is

$$y_{k+1} = y_k + \frac{\bar{k}_1 + 2\bar{k}_2 + 2\bar{k}_3 + \bar{k}_4}{6} \quad (\text{B.5})$$

where

$$\bar{k}_i^T = [\bar{k}_{1i} \ \bar{k}_{2i} \ \bar{k}_{3i} \ \dots \ \bar{k}_{ni}] \quad ; i=1 \text{ to } 4 \quad (\text{B.6})$$

Expressions for \bar{k}_1 to \bar{k}_4 are as follows:-

$$\begin{aligned} \bar{k}_1 &= hf[t_k, \bar{y}_k] \\ \bar{k}_2 &= hf\left[t_k + \frac{h}{2}, \bar{y}_k + \frac{1}{2}\bar{k}_1\right] \\ \bar{k}_3 &= hf\left[t_k + \frac{h}{2}, \bar{y}_k + \frac{1}{2}\bar{k}_2\right] \\ \bar{k}_4 &= hf[(t_k + h), (y_k + \bar{k}_3)] \end{aligned}$$

(B.7 to B.10)

2.2.8 Model by Mitchell et al. (2002)

Objective:-

- i) To analyze the performance of a Zymotis Bioreactor by investigating the optimal value for the spacing between its cooling plates.

Species: - Aspergillus niger

Substrate: - Starchy substrate

Height = 2.5 m

Superficial Air Velocity = 0.01m/s

Plate half spacing = 0.03 m

Assumptions:-

- 1) Air flows in the vertical direction, hence vertical conduction is ignored.
- 2) Void fraction does not change with time.
- 3) The system is modelled such that the heat transfer in centre is responsible for removing heat from the two half slabs of fermenting substrate.

Constitutive Relationships:-

- 1) Logistic Growth Kinetics
- 2) Density and Thermal Conductivity are calculated as volume weighted averages, whereas heat capacity is determined as a mass-weighted average of substrate bed and air properties.

Method of Solution:-

The system of model equations was non-dimensionalized and solved by method of characteristics using Orthogonal Collocation applied in both horizontal and vertical dimensions. The resulting differential algebraic system was solved by DASSL routine.

Conclusions:-

- 1) A method of productivity analysis was proposed.
- 2) Pressure drop occurring across the plate of the order of 213 Pa for a plate width of 10 mm. (Two walls of 3 mm. each and a gap width of 4 mm.) was calculated and found to be acceptable for the system considered.

2.2.9 Model of Dos Santos et al. (2004)

Objective:-

- i) To investigate the potential threat to enzyme losses caused by increased temperatures in packed bed bioreactors.

Species: - *Penicillium fellutanum*

Substrate: - Sunflower seeds

Height of Bioreactor = 1 m

Diameter of Bioreactor = 1 m

Temperature of Substrate Bed = 30°C

Superficial Velocity = 60 kg dry air/hr.

Assumptions:-

- 1) The substrate bed is well mixed, with the air leaving the bioreactor in equilibrium with the substrate bed.
- 2) The specific enzyme production rate is assumed to be unaffected by temperature. The rate constant of the denaturation reaction is expressed as a function of temperature.
- 3) Specific growth rate constant is assumed to be unaffected by changes in temperature.

Constitutive Relationships:-

- 1) Logistic Growth Kinetics
- 2) Rate constant for the enzyme denaturation reaction is given by an Arrhenius type of relationship.

Method of Solution:-

The model equations were integrated by a subroutine DRKGS within a program written in FORTRAN.

Conclusions:-

- 1) The study exemplifies that how Solid State Fermentation can be employed for studies on enzyme growth and taking into consideration phenomena pertaining to enzyme denaturation.
- 2) Simulations were conducted for *Penicillium fellutanum*, a slower growing species and *Aspergillus niger*, a faster growing species. It was concluded that the effect of cooling water played an important role in controlling the establishment of temperature gradients.

2.3 MATHEMATICAL MODELS TREATING AIR AND SUBSTRATE BED AS DISTINCT PHASES

2.3.1 Model of Weber et al. (1999)

Objective:-

- i) To explore the potential of production of conidia of the biocontrol fungus *Coniothyrium minitans* on an industrial scale.

Species: - *Coniothyrium minitans*

Substrate: - Hemp, perlite, bagasse, oats

Height of bioreactor = 1 m

Temperature of inlet air = 18°C

Superficial aeration rate = 0.013 to 0.025 kg air/m³ reactor

Assumptions:-

- 1) Radial gradients have been neglected.
- 2) Air follows ideal plug flow behaviour.

- 3) In the accumulation terms of all balances, contribution of gases and all mass accumulation terms are negligible.
- 4) Pseudosteady state with respect to temperature and oxygen consumption is assumed.
- 5) Axial gradients in oxygen consumption are neglected.
- 6) Bed porosity is constant.

Constitutive Relationships:-

- 1) Biomass production rate, water and substrate consumption rates are all related to oxygen production rates.

Conclusions:-

- 1) In this paper, a model has been developed in which a water balance has been incorporated to predict the water content of the solid substrate in a packed bed.
- 2) Oxygen consumption rate is used to estimate heat production rate which also includes the heat generated by maintenance metabolism.

2.3.2 Model of Weber et al. (2002)

Objective:-

- i) To validate the model proposed in 1999 (2.3.1) with experiments in a 15 dm³ packed bed bioreactor.

Species: - Coniothyrium minitans

Substrate: - Hemp and Oats

Diameter of the bioreactor = 20 cm.

Height of the bioreactor = 70 cm. (Bed Height = 50 cm.)

Superficial aeration rate = 0.027 – 0.069 kg/m³s

Assumptions:-

- 1) The water present in the biomass and in the substrate is not discriminated.
- 2) Air is in equilibrium with the solid matrix at any point in the bed.

iii) The denatured enzyme in all stages is

$$D_{e_n} = 0 \text{ PU/g IDS} \quad (3.36)$$

iv) Solid substrate active enzyme content is

$$E_0 = 1 \text{ PU/g IDS} \quad (3.37)$$

The model equations were solved for a 1m diameter and 1 m height bioreactor as had been done by Dos Santos et al.(2004).The air flow rate , F_a was 60 kg dry air/hr.

3.1.6 Method of Solution and Computer Program

To solve the set of simultaneous ordinary differential equations, (3.30-3.33) employing the mixed tanks-in series approach, a computer program in in MATLAB 6.1. (Math Works Inc.) was developed.

The numerical method used for the solution is Runge-Kutta-Fehlberg method (Matthews and Fink, 2004) without step size control (Appendix A).

The main inputs to the computer program are:-

1. Flow rate of air entering the bioreactor.
2. Temperature of the inlet air.
3. Solid Substrate Active Enzyme Content.
4. Number of continuous mixed flow tank reactors in which the packed bed bioreactor is visualized to be divided.
5. Denatured Enzyme Content.

The main outputs of the computer program are the profiles of the following variables with time for the visualized continuous stirred tank reactors.

1. Temperature of the bioreactor
2. Biomass production
3. Protease Enzyme Activity
4. Denaturation of Protease Enzyme

The complete computer program may be obtained from the author or from his supervisor on request.

3.2 MATHEMATICAL MODEL FOR A TWO PHASE SYSTEM

The second mathematical model which has been developed using the N CSTR's in series approach considers air(gaseous) and the dry solid bed as two distinct phases as compared to the earlier model where the two phases – gaseous and solid were modelled as a pseudohomogeneous phase.

Von Meien and Mitchell (2002) considered the growth of *Aspergillus niger* on corn wherein a non-equilibrium situation was described between the gas and solid phases. They have considered mass transfer of water between the two phases. Only Weber et al. (1999, 2002) had incorporated a water balance in his model and thus considered the resulting mass transfer effects.

3.2.1 Derivation of the Mathematical Model

The mathematical model has been derived for a cylindrical shaped packed bed reactor. The equations for two equal sized mixed tank in series system has been developed which later has been generalized for n mixed tanks in series.

3.2.2 Assumptions

The assumptions employed in the derivation of the mathematical model are:-

1. The bioreactor is assumed to be wide enough so that heat transfer to the walls makes a negligible contribution to cooling .Hence heat transfer in axial direction is considered.
2. The isotherm for corn grits has been assumed to be similar to that of the substrate.
3. The dry solid concentration and the biomass concentration do not vary in the bioreactor .

$$\text{i.e. } S_1 = S_2 = S_3 = \dots S_n = S$$

$$b_1 = b_2 = b_3 = \dots b_n = b$$

4. The reference temperature in the evaluation of convection terms(in the energy balances) is 0°C.
5. The void fraction does not change with time.
6. Effect of particle size and pressure drop has not been considered.
7. The gas flow rate per unit area does not change from bioreactor to bioreactor.

Bioreactor 2:-

$$\frac{dS_2}{dt} = Y_{SB} \frac{d(bS_2)}{dt} \quad (3.52(b))$$

$$\frac{dS_2}{dt} = Y_{SB} \left(b \frac{dS_2}{dt} + S_2 \frac{db}{dt} \right) \quad (3.53(b))$$

$$\frac{dS_2}{dt} = \frac{Y_{SB} S_2}{(1 - bY_{SB})} \frac{db}{dt} \quad (3.54(b))$$

3.2.3.5 Energy Balance for air (gas phase)

Bioreactor 1:-

Rate of energy in by convective flow of air =

$$\left(\begin{array}{c} \text{Mass Flow} \\ \text{Rate per unit} \\ \text{Cross Section} \\ \text{Area} \end{array} \right) \left(\begin{array}{c} \text{Cross} \\ \text{Section} \\ \text{Area} \end{array} \right) \left(\begin{array}{c} \text{Specific} \\ \text{Heat of} \\ \text{the} \\ \text{Water-air} \\ \text{mixture} \end{array} \right) \left(\begin{array}{c} \text{Temperature} \\ \text{Difference} \end{array} \right)$$

$$= GA(C_{pg} + \Phi_g C_{pv})(T_{g_1} - 0)$$

$$= G(C_{pg} + \Phi_g C_{pv})T_{g_1}A \quad (3.55(a))$$

$$\text{Rate of energy out by convective flow of air} = GA(C_{pg} + \Phi_{g_1} C_{pv})(T_{g_1} - 0)$$

$$= G(C_{pg} + \Phi_{g_1} C_{pv})T_{g_1}A$$

$$(3.56(a))$$

$$\text{Rate of energy accumulation in air} = \frac{1}{\Delta t} \left[\varepsilon \rho_g (C_{pg} + \Phi_{g_1} C_{pv}) \Delta T_{g_1} \right] AH_1$$

$$(3.57(a))$$

$$\text{Rate of energy transferred from air to solid phase} = h_i (T_{g_1} - T_{s_1}) AH_1$$

$$(3.58(a))$$

Formulating the energy balance for air

$$\left(\begin{array}{c} \text{Rate of} \\ \text{energy} \\ \text{accumulated} \\ \text{of water in} \\ \text{bioreactor 2} \end{array} \right) = \left(\begin{array}{c} \text{Rate of} \\ \text{energy in by} \\ \text{air through} \\ \text{convective} \\ \text{flow} \end{array} \right) - \left(\begin{array}{c} \text{Rate of energy} \\ \text{out by air} \\ \text{through} \\ \text{convective} \\ \text{flow} \end{array} \right) - \left(\begin{array}{c} \text{Rate of} \\ \text{energy} \\ \text{transferred} \\ \text{from air to} \\ \text{substrate} \\ \text{bed.} \end{array} \right)$$

$$\frac{1}{\Delta t} \left[\varepsilon \rho_g (C_{pg} + \Phi_{g1} C_{pv}) \Delta T_{g1} \right] AH_1 = G(C_{pg} + \Phi_{g1} C_{pv}) AT_{g1} - G(C_{pg} + \Phi_{g1} C_{pv}) AT_{g1} - h_1 (T_{g1} - T_{s1}) AH_1 \quad (3.59(a))$$

Dividing by AH_1 and taking $Lt \Delta t \rightarrow 0$

$$\varepsilon \rho_g \left[(C_{pg} + \Phi_{g1} C_{pv}) \frac{dT_{g1}}{dt} \right] = G(C_{pg} + \Phi_{g1} C_{pv}) \left[\frac{T_{g1} - T_{g1}}{H_1} \right] - h_1 (T_{g1} - T_{s1}) \quad (3.60(a))$$

Bioreactor 2:-

Rate of energy in by convective flow of air =

$$\left(\begin{array}{c} \text{Mass Flow} \\ \text{Rate per unit} \\ \text{Cross Section} \\ \text{Area} \end{array} \right) \left(\begin{array}{c} \text{Cross} \\ \text{Section} \\ \text{Area} \end{array} \right) \left(\begin{array}{c} \text{Specific} \\ \text{Heat of} \\ \text{the} \\ \text{Water-air} \\ \text{mixture} \end{array} \right) \left(\begin{array}{c} \text{Temperature} \\ \text{Difference} \end{array} \right)$$

$$= GA(C_{pg} + \Phi_{g2} C_{pv})(T_{g1} - 0)$$

$$= G(C_{pg} + \Phi_{g2} C_{pv}) T_{g1} A \quad (3.55(b))$$

Rate of energy out by convective flow of air = $GA(C_{pg} + \Phi_{g2} C_{pv})(T_{g2} - 0)$

$$= G(C_{pg} + \Phi_{g2} C_{pv}) T_{g2} A \quad (3.56(b))$$

Rate of energy accumulation in air = $\frac{1}{\Delta t} \left[\varepsilon \rho_g (C_{pg} + \Phi_{g2} C_{pv}) \Delta T_{g2} \right] AH_2$

$$(3.57(b))$$

$$\text{Rate of energy transferred from air to solid phase} = h_i(T_{g_2} - T_{s_2})AH_2 \quad (3.58(b))$$

Formulating the energy balance for air

$$\left(\begin{array}{c} \text{Rate of} \\ \text{energy} \\ \text{accumulated} \\ \text{of water in} \\ \text{bioreactor 2} \end{array} \right) = \left(\begin{array}{c} \text{Rate of} \\ \text{energy in by} \\ \text{air through} \\ \text{convective} \\ \text{flow} \end{array} \right) - \left(\begin{array}{c} \text{Rate of energy} \\ \text{out by air} \\ \text{through} \\ \text{convective} \\ \text{flow} \end{array} \right) - \left(\begin{array}{c} \text{Rate of} \\ \text{energy} \\ \text{transferred} \\ \text{from air to} \\ \text{solids} \end{array} \right)$$

$$\frac{1}{\Delta t} \left[\varepsilon \rho_g (C_{pg} + \Phi_{g_2} C_{pv}) \Delta T_{g_2} \right] AH_2 = G(C_{pg} + \Phi_{g_2} C_{pv}) AT_{g_1} - G(C_{pg} + \Phi_{g_2} C_{pv}) AT_{g_2} - h_i(T_{g_2} - T_{s_2}) AH_2 \quad (3.59(b))$$

Dividing by AH_2 and taking $\text{Lt } \Delta t \rightarrow 0$

$$\varepsilon \rho_g \left[(C_{pg} + \Phi_{g_2} C_{pv}) \frac{dT_{g_2}}{dt} \right] = G(C_{pg} + \Phi_{g_2} C_{pv}) \left[\frac{T_{g_1} - T_{g_2}}{H_2} \right] - h_i(T_{g_2} - T_{s_2}) \quad (3.60(b))$$

3.2.3.6 Energy Balance for the Solid phase

Bioreactor 1:-

Rate of energy accumulation in the solid phase

$$= \frac{1}{\Delta t} S_1 (C_{ps} + \Phi_{s_1} C_{pv}) \Delta T_{s_1} (AH_1) \quad (3.61(a))$$

Rate of energy transferred due to evaporation of water from the solid phase

$$= \lambda K_a (\Phi_{s_1} - \Phi_{s_1}^*) AH_1 \quad (3.62(a))$$

Rate of energy transferred from gas to substrate bed

$$= h_i AH_1 (T_{g_1} - T_{s_1}) \quad (3.63(a))$$

Rate of energy generated by biomass production

$$= Y_Q \frac{\Delta(bS_1)}{\Delta t} AH_1 \quad (3.64(a))$$

Formulation of energy balance on the substrate bed yields:-

$$\left(\begin{array}{c} \text{Rate of energy} \\ \text{accumulated in} \\ \text{the solid phase in} \\ \text{Bioreactor 1} \end{array} \right) = \left(\begin{array}{c} \text{Rate of} \\ \text{energy} \\ \text{transferred} \\ \text{from gas to} \\ \text{solid phase} \end{array} \right) -$$

$$\left(\begin{array}{c} \text{Rate of energy} \\ \text{transferred due} \\ \text{to evaporation} \\ \text{of water from} \\ \text{solid phase} \end{array} \right) + \left(\begin{array}{c} \text{Rate of} \\ \text{energy} \\ \text{generated} \\ \text{by biomass} \\ \text{production} \end{array} \right)$$

$$\begin{aligned} \frac{1}{\Delta t} S_1 (C_{pv} + \Phi_{S_1} C_{pw}) \Delta T_{S_1} (AH_1) &= h_t AH_1 (T_{g_1} - T_{S_1}) - \lambda K_a (\Phi_{S_1} - \Phi_{S_1}^*) AH_1 \\ + Y_Q \frac{\Delta(bS_1)}{\Delta t} AH_1 & \end{aligned} \quad (3.65(a))$$

Dividing all the terms by AH_1 and taking the $Lt \Delta t \rightarrow 0$

$$\begin{aligned} S_1 (C_{pv} + \Phi_{S_1} C_{pw}) \frac{dT_{S_1}}{dt} &= h_t (T_{g_1} - T_{S_1}) - \lambda K_a (\Phi_{S_1} - \Phi_{S_1}^*) \\ + Y_Q \frac{d(bS_1)}{dt} & \end{aligned} \quad (3.66(a))$$

$$\begin{aligned} S_1 (C_{pv} + \Phi_{S_1} C_{pw}) \frac{dT_{S_1}}{dt} &= h_t (T_{g_1} - T_{S_1}) - \lambda K_a (\Phi_{S_1} - \Phi_{S_1}^*) \\ + Y_Q \left(S_1 \frac{db}{dt} + b \frac{dS_1}{dt} \right) & \end{aligned} \quad (3.67(a))$$